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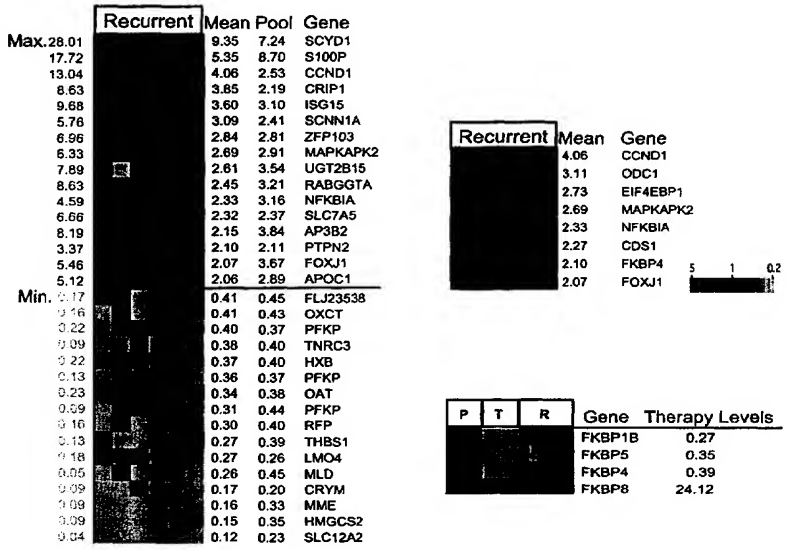
(54) Title: GENES RELATED TO DEVELOPMENT OF REFRACTORY PROSTATE CANCER



(57) Abstract: The present disclosure provides hormone-refractory prostate cancer (HPRC)-related nucleic acid molecules and proteins useful for the detection of neoplasms, particularly prostate and more specifically hormone-refractory prostate cancers. Also provided are methods of using these biological materials in the diagnosis, staging, detection, and treatment of neoplasia, and particularly hormone-refractory prostate cancer.

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GENES RELATED TO DEVELOPMENT OF REFRACTORY PROSTATE CANCER

FIELD

The present disclosure is generally related to diagnosing, prognosing, staging, preventing, and treating disease, particularly hormone refractory prostate cancer.

BACKGROUND

About 40 years ago it was observed that prostate cancer, like normal prostate, is often androgen dependent and androgen withdrawal induces growth regression. Various modes of androgen ablation therapy have since been used as the major treatment of advanced prostate cancer. Generally, chemical or physical reduction in serum androgens, or chemical blockage of their action, effectively arrests growth of local and metastatic prostate cancer *in vivo*. Unfortunately, in almost all patients, the regressed tumors eventually develop resistance to hormonal therapy and recur as aggressive androgen independent tumors that are hormone refractory and currently incurable. Identification of the genes that regulate the therapeutically induced tumor regression, and the genes associated with resistance to therapy, are candidate targets that can be used for rational design of therapeutic interventions.

The clinical course of hormone therapy response and eventual recurrence can be modeled experimentally using CWR22 xenografts. CWR22 is an androgen dependent human prostate carcinoma that grows rapidly as a xenograft in male nude mice, regresses after castration, and eventually (in three to ten months) becomes recurrent and re-grows independently of androgens in castrated mice. Several groups have previously looked for differences in gene expression between the primary and recurrent CWR22 prostate cancer xenografts, and identified some candidate genes that can be used as biomarkers. It remains critical to make sure that findings from model systems are applicable in the clinical situation.

Molecular mechanisms involved in the regression of prostate cancer after androgen deprivation, as well as in the re-growth of androgen-independent tumors, remain poorly understood. There is a need to better understand patterns of gene expression that trigger prostate tumor regression and/or re-growth, as well as downstream genes that may serve as indicators of prostate cancer progression.

BRIEF SUMMARY OF THE DISCLOSURE

Embodiments of this disclosure provide a set of nucleic acid molecules the expression of which is altered in prostate cancer, more particularly nucleic acid molecules that show temporal expression changes during prostate cancer hormonal therapy and regression.

Provided herein in various embodiments are hormone-refractory prostate cancer (HPRC)-related nucleic acid molecules and polypeptides useful for the detection/diagnosis/staging and treatment of neoplasms, particularly prostate and more specifically hormone-refractory prostate

cancers. Also provided are methods of using these biological materials in the diagnosis, staging, detection, and treatment of neoplasia, and particularly hormone-refractory prostate cancer.

The foregoing and other features and advantages of these and other embodiments will become more apparent from the following detailed description of several embodiments, which proceeds with reference to the accompanying figures.

BRIEF DESCRIPTION OF THE FIGURES

FIG 1A, 1B, 1C, and 1D are schematic diagrams that show relative expression and expression patterns of 2333 genes measured using cDNA microarray technology at various time points during prostate cancer development.

FIG 2 shows a scatter plot that was generated for one primary and one recurrent tumor. This plot demonstrates the correlation between the samples (low variance, as indicated by lack of scatter in the majority of the genes), and highlights the most differentially expressed genes (which are circled and named).

FIG 3 shows lists of genes that were differentially expressed between primary and recurrent xenografts, along with related relative expression information. A set of 30 genes were most consistently differentially expressed (out of a total of 164 genes that changed 2-fold or more) in two independent experiments.

FIG. 3A shows the 30 genes, ordered by degree of differential expression. The grey-shade coding reflects the relative gene expression ratio (normalized to the mean ratio for four primary tumors) for each of six different recurrent xenografts tumors (arranged in columns). For the six recurrent tumors, the mean expression ratio relative to the mean expression levels of the primary tumors is indicated in the "Mean" column. Additionally, the maximum ratio (Max.) for the upregulated genes, and the minimum ratio (Min.) for the downregulated genes is also indicated (left column). The "pool" column depicts the ratios of a direct cDNA microarray experiment where four primary tumors were pooled and compared to four recurrent tumors.

FIG. 3B shows eight PI3/AKT/FRAP pathway-related genes, the expression of which was associated with hormone-refractory cell growth (based on >two-fold induction of in the recurrent tumors relative to the primary level). Grey-shade-coded gene expression ratios as well as the mean are shown as in FIG. 3A. The criteria for selecting these genes were i) a >two-fold change in the average ratio between primary and recurrent tumors (or during therapy) and ii) evidence from the literature suggesting the interaction of these gene products with macrolide drugs or their involvement in a rapamycin-sensitive pathway.

FIG. 3C shows four FK506-binding protein genes, which were associated with hormone refractory tumor growth (based on at least a two-fold response to therapy and were restored to greater than 80 % of primary levels in the recurrent tumors). Grey-shade-coded gene expression ratios are shown for each of four primary (P) tumors, four tumors regressing following therapy (T), and six

recurrent tumors (R). The mean ratio of gene expression (relative to the primary tumors) is shown for tumors undergoing therapy.

FIG. 3D is a graph showing cell viability after treatment with Rapamycin (solid lines) and FK506 (dotted lines). The effects of these drugs on the viability of the hormone refractory CWR22R cell line (marked with a "C") and LNCap (marked with an "L") was tested *in vitro*. The recurrent CWR22R cell line was highly sensitive to rapamycin ($IC_{50} \sim 0.1$ nM) and underwent cell death. In contrast, LNCap showed partial growth arrest without cell death, even at higher doses of rapamycin. FK506 did not have an effect on either cell line. ED_{50} in CWR22R cells for rRapamycin was 0.3 μM.

FIG 4 shows S100P mRNA levels measured by three different methods (cDNA microarray, mRNA ISH, and Northern hybridization analyses) in nine xenografts. The amount of S100P detected in each of these three methods was quantified and plotted in a line graph above the corresponding images. Absolute values were normalized to the three primary tumors with the lowest Northern hybridization levels.

FIG 5 is a bar graph showing the level of S100P protein expression in 440 human prostate cancer specimens at various stages of progression, measured by IHC staining. An S100P antibody was used to stain prostate tissue sections on a tissue microarray containing hundreds of prostate specimens from different steps of cancer development (from normal epithelium, BPH, and localized cancer to metastases and hormone refractory prostate cancer). The staining was scored by two pathologists, using a scale of 0 to 4. The results show the percentage of cancers at each progression stage that had strong (score of 3 or 4) IHC staining.

FIG 6 shows the results of analyses of specific gene targets involved in drug response. The top graphs (FIGs 6A and 6B) illustrate the dose response of CWR22R cell line viability *in vitro* with various emerging therapies (TSA, FR901464, rapamycin, RSD, FK506, and androgen withdrawal therapy); the levels of FKBP5 (FIG 6A) and VDUP1 (FIG 6B) are shown. A time course of treatment for each drug was analyzed by cDNA microarray and a database of the resulting data was mined to find genes that are involved in more than one therapeutic response. Specific examples are shown, including CRYM ATP1B2, OAT, QSCN6, GSN, PLU-1, GFPT2, ZCYTOR7, and VDUP1.

FIG 6C shows representative quantitative analyses for expression of the indicated genes at 0, 1, 3, 9, and 24 hours after treatment with the indicated drugs (0.3 μM TSA, 10 mM FR901464, 1μM rapamycin, 1μM FK506, and 1 μM RSD).

FIG 6D shows the expression levels for the same genes in primary, regressing, and recurrent tumors.

SEQUENCE LISTING

The nucleic and amino acid sequences listed in the accompanying sequence listing are shown using standard letter abbreviations for nucleotide bases, and three letter code for amino acids, as defined in 37 C.F.R. 1.822. Only one strand of each nucleic acid sequence is shown, but the complementary strand is understood as included by any reference to the displayed strand. In the accompanying sequence listing:

SEQ ID NOs: 1 and 2, also referred to as S100PF and S100PR respectively, are examples of oligonucleotides useful for amplifying an S100p probe sequence.

SEQ ID NOs: 3-10 (AntiS100P-A, -B, -C, -D, -E, -F, -G, and –H respectively, are examples of oligonucleotides useful for mRNA *in situ* hybridization.

DETAILED DESCRIPTION

I. Abbreviations

AR	androgen receptor
LNCap	prostate cancer cell line (developed by Dr. Leland Chung)
HRPC	hormone refractory prostate cancer
ED50	50% Effective Dose
ISH	*in situ* hybridization
IHC	immunohistochemical
MTT	(3-(4,5-dimethyl-2-thiazolyl)-2,5-diphenyl-2H-tetrazolium bromide)
TSA	trichostatin A
PCNA	proliferating cell nuclear antigen

II. Terms

Unless otherwise noted, technical terms are used according to conventional usage. Definitions of common terms in molecular biology may be found in Benjamin Lewin, *Genes V*, published by Oxford University Press, 1994 (ISBN 0-19-854287-9); Kendrew *et al.* (eds.), *The Encyclopedia of Molecular Biology*, published by Blackwell Science Ltd., 1994 (ISBN 0-632-02182-9); and Robert A. Meyers (ed.), *Molecular Biology and Biotechnology: a Comprehensive Desk Reference*, published by VCH Publishers, Inc., 1995 (ISBN 1-56081-569-8).

In order to facilitate review of the various embodiments, the following explanations of certain terms are provided:

Analog, derivative or mimetic: An analog is a molecule that differs in chemical structure from a parent compound, for example a homolog (differing by an increment in the chemical structure, such as a difference in the length of an alkyl chain, though the change need not be an incremental change in length of a chain), a molecular fragment, a structure that differs by one or more functional groups, a change in ionization. Structural analogs are often found using quantitative structure activity

relationships (QSAR), with techniques such as those disclosed in Remington (*The Science and Practice of Pharmacology*, 19th Edition (1995), chapter 28). A derivative is a biologically active molecule derived from the base (parental) structure. A mimetic is a biomolecule that mimics the activity of another biologically active molecule.

Biologically active molecules can include chemical structures that mimic the biological activities of a compound, for instance rapamycin or more generally macrolides (the basic tri-cyclic structural group that includes rapamycin). Rapamycin derivatives (including metabolic derivatives), analogs, and mimetics are disclosed, for instance, in USPN 5,508,398; Kuhn *et al.*, *J. Med. Chem.* 44:2027-2034, 2001; Dickman *et al.*, Bioorg. Med. Chem. Lett 10:1405-1408, 2000; Streit *et al.*, Drug Metab. Dispos. 24:1272-1278, 1996; and Wong *et al.*, *J. Antibiot. (Tokyo)* 51:487-491, 1998.

Antisense, Sense, and Antigene: Double-stranded DNA (dsDNA) has two strands, a 5' -> 3' strand, referred to as the plus strand, and a 3' -> 5' strand (the reverse complement), referred to as the minus strand. Because RNA polymerase adds nucleic acids in a 5' -> 3' direction, the minus strand of the DNA serves as the template for the RNA during transcription. Thus, the RNA formed will have a sequence complementary to the minus strand and identical to the plus strand (except that U is substituted for T).

Antisense molecules are molecules that are specifically hybridizable or specifically complementary to either RNA or the plus strand of DNA. Sense molecules are molecules that are specifically hybridizable or specifically complementary to the minus strand of DNA. Antigene molecules are either antisense or sense molecules directed to a dsDNA target.

A gene-suppressive technology that is similar to antisense technology involves the use of small inhibitory RNA molecules (siRNAs) to inhibit a target gene. Methods of using siRNAs to inhibit eukaryotic and more particularly mammalian gene expression are known to those of ordinary skill in the art; see, for instance, Caplen *et al.*, *Proc. Natl. Acad. Sci.* 98(17):9742-9747, 2001, and Elbashir *et al.*, *Nature* 411:494-498, 2001.

Array: An arrangement of molecules, particularly biological macromolecules (such as polypeptides or nucleic acids) or cell or tissue samples, in addressable locations on or in a substrate. The array may be regular (arranged in uniform rows and columns, for instance) or irregular. The number of addressable locations on the array can vary, for example from a few (such as three) to more than 50, 100, 200, 500, 1000, 10,000, or more. A "microarray" is an array that is miniaturized so as to require or be aided by microscopic examination for evaluation or analysis.

Within an array, each arrayed sample (feature) is addressable, in that its location can be reliably and consistently determined within the at least two dimensions of the array. Thus, in ordered arrays the location of each sample is assigned to the sample at the time when it is applied to the array, and a key may be provided in order to correlate each location with the appropriate target or feature position. Often, ordered arrays are arranged in a symmetrical grid pattern, but samples could be arranged in other patterns (*e.g.*, in radially distributed lines, spiral lines, or ordered clusters). Addressable arrays usually are computer readable, in that a computer can be programmed to correlate

a particular address on the array with information about the sample at that position (*e.g.*, hybridization or binding data, including for instance signal intensity). In some examples of computer readable formats, the individual features in the array are arranged regularly, for instance in a Cartesian grid pattern, which can be correlated to address information by a computer.

The sample application location on an array (the "feature") may assume many different shapes. Thus, though the term "spot" may be used herein, it refers generally to a localized placement of molecules or tissue or cells, and is not limited to a round or substantially round region. For instance, substantially square regions of application can be used with arrays encompassed herein, as can be regions that are, for example substantially rectangular, triangular, oval, irregular, or another shape.

In certain example arrays, one or more features will occur on the array a plurality of times (*e.g.*, twice) to provide internal controls.

Binding or stable binding: An oligonucleotide binds or stably binds to a target nucleic acid if a sufficient amount of the oligonucleotide forms base pairs or is hybridized to its target nucleic acid, to permit detection of that binding. Binding can be detected by either physical or functional properties of the target:oligonucleotide complex. Binding between a target and an oligonucleotide can be detected by any procedure known to one skilled in the art, including both functional and physical binding assays. Binding may be detected functionally by determining whether binding has an observable effect upon a biosynthetic process such as expression of a gene, DNA replication, transcription, translation, and the like.

Physical methods of detecting the binding of complementary strands of DNA or RNA are well known in the art, and include such methods as DNase I or chemical footprinting, gel shift and affinity cleavage assays, Northern blotting, dot blotting and light absorption detection procedures. For example, one method that is widely used, because it is so simple and reliable, involves observing a change in light absorption of a solution containing an oligonucleotide (or an analog) and a target nucleic acid at 220 to 300 nm as the temperature is slowly increased. If the oligonucleotide or analog has bound to its target, there is a sudden increase in absorption at a characteristic temperature as the oligonucleotide (or analog) and target disassociate from each other, or melt.

The binding between an oligomer and its target nucleic acid is frequently characterized by the temperature (T_m) at which 50% of the oligomer is melted from its target. A higher (T_m) means a stronger or more stable complex relative to a complex with a lower (T_m).

Cancer: A cancer is a biological condition in which a malignant tumor or other neoplasm has undergone characteristic anaplasia with loss of differentiation, increased rate of growth, invasion of surrounding tissue, and which is capable of metastasis.

The term cancer includes prostate cancer, such as prostate adenocarcinoma, transitional cell carcinomas, squamous cell carcinomas, and sarcomas. However, about 95% of prostate cancers are adenocarcinomas. Also included are different stages of a single cancer, for instance both primary and recurrent (hormone-refractory) prostate cancer.

cDNA (complementary DNA): A piece of DNA lacking internal, non-coding segments (introns) and transcriptional regulatory sequences. cDNA may also contain untranslated regions (UTRs) that are responsible for translational control in the corresponding RNA molecule. cDNA is usually synthesized in the laboratory by reverse transcription from messenger RNA extracted from cells.

Comparative genomic hybridization: A technique of differential labeling of test DNA and normal reference DNA, which are hybridized simultaneously to chromosome spreads, as described in Kallioniemi *et al.* (*Science* 258:818-821, 1992), incorporated by reference.

Complementarity and percentage complementarity: Molecules with complementary nucleic acids form a stable duplex or triplex when the strands bind, (hybridize), to each other by forming Watson-Crick, Hoogsteen or reverse Hoogsteen base pairs. Stable binding occurs when an oligonucleotide remains detectably bound to a target nucleic acid sequence under the required conditions.

Complementarity is the degree to which bases in one nucleic acid strand base pair with the bases in a second nucleic acid strand. Complementarity is conveniently described by percentage, *i.e.* the proportion of nucleotides that form base pairs between two strands or within a specific region or domain of two strands. For example, if 10 nucleotides of a 15-nucleotide oligonucleotide form base pairs with a targeted region of a DNA molecule, that oligonucleotide is said to have 66.67% complementarity to the region of DNA targeted.

In the present disclosure, "sufficient complementarity" means that a sufficient number of base pairs exist between the oligonucleotide and the target sequence to achieve detectable binding, and in the case of the binding of an antigen, disrupt expression of gene products (such as cartilage glycoprotein-39 (CHI3L1), S-100P PROTEIN (S100P), CX3C chemokine/fractalkine (SCYD1), adenylate kinase 1 (AK1), forkhead transcription factor HFH-4 (HFH-4) (FKHL13), UDP glucuronosyltransferase precursor (UGT2B15) , Pleiotrophin (heparin binding growth factor 8) (PTN), heat shock 27kD protein 2/Alpha-B-crystallin (HSP27), Proteasome (prosome, macropain) subunit, beta type, 5 (PSMB5), Inhibitor of NFKB (NFKBIA), interferon-induced 17 kD protein (ISG15), MAP kinase activated protein kinase 2 (MAPKAPK2), signal transduction protein (SH3 containing) (EFS2), hkf-1 Zinc finger protein (ZFP103), chromosome condensation 1 (CHC1), CDP-diacylglycerol synthase (CDS1), gap junction protein, alpha 1, 43kD (connexin 43) (GJA1), cyclin D1 (CCND1), Inhibitor of DNA binding 3, helix-loop-helix protein (ID3), H1 histone family, member2 (H1F2), Cytochrome B561 (CYB561), Cathepsin H (CTSH), calcineurin alpha (PPP3CA), 54 kDa progesterone receptor-associated immunophilin (FKBP5), translocation protein 1 (TLOC1), Clusterin (complement lysis inhibitor; testosterone-repressed prostate message 2; apolipoprotein J) (CLU), Pulmonary surfactant-associated protein A (SFTPA1), protease inhibitor 12 (PI12; neuroserpin) (PI12), Thrombospondin 1 (THBS1), Ribophorin I (RPN1), A disintegrin-like and metalloprotease (reprolysin type) with thrombospondin type 1 motif, 1 (ADAMTS1), Collagen, type IV, alpha 5 (Alport syndrome) (COL4A5), LIM domain only 4 / breast tumor autoantigen (LMO4),

bumetanide-sensitive Na-K-Cl cotransporter (NKCC1) (SLC12A2), Fibronectin (FN1), Crystallin Mu (CRYM) and "upregulated by 1,25-dihydroxyvitamin D-3" (VDUP1)). When expressed or measured by percentage of base pairs formed, the percentage complementarity that fulfills this goal can range from as little as about 50% complementarity to full (100%) complementary. In general, sufficient complementarity is at least about 50%, about 75% complementarity, about 90% or 95% complementarity, and or about 98% or even 100% complementarity.

A thorough treatment of the qualitative and quantitative considerations involved in establishing binding conditions that allow one skilled in the art to design appropriate oligonucleotides for use under the desired conditions is provided by Beltz *et al. Methods Enzymol* 100:266-285, 1983, and by Sambrook *et al.* (ed.), *Molecular Cloning: A Laboratory Manual*, 2nd ed., vol. 1-3, Cold Spring Harbor Laboratory Press, Cold Spring Harbor, NY, 1989.

DNA (deoxyribonucleic acid): DNA is a long chain polymer which comprises the genetic material of most living organisms (some viruses have genes comprising ribonucleic acid (RNA)). The repeating units in DNA polymers are four different nucleotides, each of which comprises one of the four bases, adenine, guanine, cytosine and thymine bound to a deoxyribose sugar to which a phosphate group is attached. Triplets of nucleotides (referred to as codons) code for each amino acid in a polypeptide, or for a stop signal. The term codon is also used for the corresponding (and complementary) sequences of three nucleotides in the mRNA into which the DNA sequence is transcribed.

Unless otherwise specified, any reference to a DNA molecule is intended to include the reverse complement of that DNA molecule. Except where single-strandedness is required by the text herein, DNA molecules, though written to depict only a single strand, encompass both strands of a double-stranded DNA molecule. Thus, a reference to the nucleic acid molecule that encodes a specific protein, or a fragment thereof, encompasses both the sense strand and its reverse complement. Thus, for instance, it is appropriate to generate probes or primers from the reverse complement sequence of the disclosed nucleic acid molecules.

Deletion: The removal of a sequence of DNA, the regions on either side of the removed sequence being joined together.

Gene amplification or **genomic amplification**: An increase in the copy number of a gene or a fragment or region of a gene or associated 5' or 3' region, as compared to the copy number in normal tissue. An example of a genomic amplification is an increase in the copy number of an oncogene. A "gene deletion" is a deletion of one or more nucleic acids normally present in a gene sequence and, in extreme examples, can include deletions of entire genes or even portions of chromosomes.

Gene expression fingerprint (or profile): A distinct or identifiable pattern of gene expression, for instance a pattern of high and low expression of a defined set of genes; in some instances, as few as one or two genes may provide a profile, but often more genes are used in a profile, for instance at least three, at least 5, at least 10, at least 20, at least 25, or at least 50 or more.

Gene expression fingerprints (also referred to as profiles) can be linked to a tissue or cell type, to a particular stage of normal tissue growth or disease progression, or to any other distinct or identifiable condition that influences gene expression in a predictable way. Gene expression fingerprints can include relative as well as absolute expression levels of specific genes, and often are best viewed in the context of a test sample compared to a baseline or control sample fingerprint. By way of example, a gene expression profile may be read on an array (*e.g.*, a polynucleotide or polypeptide array). Arrays are now well known, and for instance gene expression arrays have been previously described in published PCT application number US99/06860 ("Hypoxia-Inducible Human Genes, Proteins, and Uses Thereof"), incorporated herein by reference in its entirety.

Genomic target sequence: A sequence of nucleotides located in a particular region in the human genome that corresponds to one or more specific genetic abnormalities, such as a nucleotide polymorphism, a deletion, or an amplification. The target can be for instance a coding sequence; it can also be the non-coding strand that corresponds to a coding sequence.

HRPC-related molecule: A molecule that is involved in, or influenced by, hormone-refractory prostate cancer. Such molecules include, for instance, nucleic acids (*e.g.*, DNA, cDNA, or mRNAs) and proteins. Specific examples of HRPC-related molecules include the nucleic acid molecules listed in Table 1, and proteins or protein fragments encoded thereby. HRPC-related molecules may be involved in or influenced by hormone-refractory prostate cancer in many different ways, including causative (in that a change in an HRPC-related molecule leads to development of or progression to hormone-refractory prostate cancer) or resultive (in that development of or progression to hormone-refractory prostate cancer causes or results in a change in the HRPC-related molecule).

Hybridization: Oligonucleotides and their analogs hybridize by hydrogen bonding, which includes Watson-Crick, Hoogsteen or reversed Hoogsteen hydrogen bonding, between complementary bases. Generally, nucleic acid consists of nitrogenous bases that are either pyrimidines (cytosine (C), uracil (U), and thymine (T)) or purines (adenine (A) and guanine (G)). These nitrogenous bases form hydrogen bonds between a pyrimidine and a purine, and the bonding of the pyrimidine to the purine is referred to as "base pairing." More specifically, A will hydrogen bond to T or U, and G will bond to C. "Complementary" refers to the base pairing that occurs between to distinct nucleic acid sequences or two distinct regions of the same nucleic acid sequence.

"Specifically hybridizable" and "specifically complementary" are terms that indicate a sufficient degree of complementarity such that stable and specific binding occurs between the oligonucleotide (or its analog) and the DNA or RNA target. The oligonucleotide or oligonucleotide analog need not be 100% complementary to its target sequence to be specifically hybridizable. An oligonucleotide or analog is specifically hybridizable when binding of the oligonucleotide or analog to the target DNA or RNA molecule interferes with the normal function of the target DNA or RNA, and there is a sufficient degree of complementarity to avoid non-specific binding of the oligonucleotide or analog to non-target sequences under conditions where specific binding is desired,

for example under physiological conditions in the case of *in vivo* assays or systems. Such binding is referred to as specific hybridization.

Hybridization conditions resulting in particular degrees of stringency will vary depending upon the nature of the hybridization method of choice and the composition and length of the hybridizing nucleic acid sequences. Generally, the temperature of hybridization and the ionic strength (especially the Na^+ concentration) of the hybridization buffer will determine the stringency of hybridization, though waste times also influence stringency. Calculations regarding hybridization conditions required for attaining particular degrees of stringency are discussed by Sambrook *et al.* (ed.), *Molecular Cloning: A Laboratory Manual*, 2nd ed., vol. 1-3, Cold Spring Harbor Laboratory Press, Cold Spring Harbor, NY, 1989, chapters 9 and 11, herein incorporated by reference.

For purposes of the present disclosure, "stringent conditions" encompass conditions under which hybridization will only occur if there is less than 25% mismatch between the hybridization molecule and the target sequence. "Stringent conditions" may be broken down into particular levels of stringency for more precise definition. Thus, as used herein, "moderate stringency" conditions are those under which molecules with more than 25% sequence mismatch will not hybridize; conditions of "medium stringency" are those under which molecules with more than 15% mismatch will not hybridize, and conditions of "high stringency" are those under which sequences with more than 10% mismatch will not hybridize. Conditions of "very high stringency" are those under which sequences with more than 6% mismatch will not hybridize.

***In vitro* amplification:** Techniques that increase the number of copies of a nucleic acid molecule in a sample or specimen. An example of amplification is the polymerase chain reaction, in which a biological sample collected from a subject is contacted with a pair of oligonucleotide primers, under conditions that allow for the hybridization of the primers to nucleic acid template in the sample. The primers are extended under suitable conditions, dissociated from the template, and then re-annealed, extended, and dissociated to amplify the number of copies of the nucleic acid. The product of *in vitro* amplification may be characterized by electrophoresis, restriction endonuclease cleavage patterns, oligonucleotide hybridization or ligation, and/or nucleic acid sequencing, using standard techniques. Other examples of *in vitro* amplification techniques include strand displacement amplification (see U.S. Patent No. 5,744,311); transcription-free isothermal amplification (see U.S. Patent No. 6,033,881); repair chain reaction amplification (see WO 90/01069); ligase chain reaction amplification (see EP-A-320 308); gap filling ligase chain reaction amplification (see U.S. Patent No. 5,427,930); coupled ligase detection and PCR (see U.S. Patent No. 6,027,889); and NASBA™ RNA transcription-free amplification (see U.S. Patent No. 6,025,134).

Injectable composition: A pharmaceutically acceptable fluid composition including at least one active ingredient. The active ingredient is usually dissolved or suspended in a physiologically acceptable carrier, and the composition can additionally include minor amounts of one or more non-toxic auxiliary substances, such as emulsifying agents, preservatives, and pH buffering agents and the like. Such injectable compositions that are useful for use with the

nucleotides and proteins of this disclosure are conventional; appropriate formulations are well known in the art.

Isolated: An "isolated" biological component (such as a nucleic acid molecule, protein or organelle) has been substantially separated or purified away from other biological components in the cell of the organism in which the component naturally occurs, *i.e.*, other chromosomal and extra-chromosomal DNA and RNA, proteins and organelles. Nucleic acids and proteins that have been "isolated" include nucleic acids and proteins purified by standard purification methods. The term also embraces nucleic acids and proteins prepared by recombinant expression in a host cell as well as chemically synthesized nucleic acids.

Neoplasm: A new and abnormal growth, particularly a new growth of tissue or cells in which the growth is uncontrolled and progressive. A tumor is an example of a neoplasm.

Nucleotide: "Nucleotide" includes, but is not limited to, a monomer that includes a base linked to a sugar, such as a pyrimidine, purine or synthetic analogs thereof, or a base linked to an amino acid, as in a peptide nucleic acid (PNA). A nucleotide is one monomer in a polynucleotide. A nucleotide sequence refers to the sequence of bases in a polynucleotide.

Oligonucleotide: An oligonucleotide is a plurality of joined nucleotides joined by native phosphodiester bonds, between about 6 and about 300 nucleotides in length. An oligonucleotide analog refers to moieties that function similarly to oligonucleotides but have non-naturally occurring portions. For example, oligonucleotide analogs can contain non-naturally occurring portions, such as altered sugar moieties or inter-sugar linkages, such as a phosphorothioate oligodeoxynucleotide. Functional analogs of naturally occurring polynucleotides can bind to RNA or DNA, and include peptide nucleic acid (PNA) molecules.

Particular oligonucleotides and oligonucleotide analogs can include linear sequences up to about 200 nucleotides in length, for example a sequence (such as DNA or RNA) that is at least 6 bases, for example at least 8, 10, 15, 20, 25, 30, 35, 40, 45, 50, 100 or even 200 bases long, or from about 6 to about 50 bases, for example about 10-25 bases, such as 12, 15 or 20 bases.

Operably linked: A first nucleic acid sequence is operably linked with a second nucleic acid sequence when the first nucleic acid sequence is placed in a functional relationship with the second nucleic acid sequence. For instance, a promoter is operably linked to a coding sequence if the promoter affects the transcription or expression of the coding sequence. Generally, operably linked DNA sequences are contiguous and, where necessary to join two protein-coding regions, in the same reading frame.

Open reading frame: A series of nucleotide triplets (codons) coding for amino acids without any internal termination codons. These sequences are usually translatable into a peptide.

Ortholog: Two nucleic acid or amino acid sequences are orthologs of each other if they share a common ancestral sequence and diverged when a species carrying that ancestral sequence split into two species. Orthologous sequences are also homologous sequences.

Parenteral: Administered outside of the intestine, *e.g.*, not via the alimentary tract. Generally, parenteral formulations are those that will be administered through any possible mode except ingestion. This term especially refers to injections, whether administered intravenously, intrathecally, intramuscularly, intraperitoneally, or subcutaneously, and various surface applications including intranasal, intradermal, and topical application, for instance.

Peptide Nucleic Acid (PNA): An oligonucleotide analog with a backbone comprised of monomers coupled by amide (peptide) bonds, such as amino acid monomers joined by peptide bonds.

Pharmaceutically acceptable carriers: The pharmaceutically acceptable carriers useful in this disclosure are conventional. Martin, *Remington's Pharmaceutical Sciences*, published by Mack Publishing Co., Easton, PA, 19th Edition, 1995, describes compositions and formulations suitable for pharmaceutical delivery of the nucleotides and proteins herein disclosed.

In general, the nature of the carrier will depend on the particular mode of administration being employed. For instance, parenteral formulations usually comprise injectable fluids that include pharmaceutically and physiologically acceptable fluids such as water, physiological saline, balanced salt solutions, aqueous dextrose, glycerol or the like as a vehicle. For solid compositions (*e.g.*, powder, pill, tablet, or capsule forms), conventional non-toxic solid carriers can include, for example, pharmaceutical grades of mannitol, lactose, starch, or magnesium stearate. In addition to biologically-neutral carriers, pharmaceutical compositions to be administered can contain minor amounts of non-toxic auxiliary substances, such as wetting or emulsifying agents, preservatives, and pH buffering agents and the like, for example sodium acetate or sorbitan monolaurate.

Probes and primers: Nucleic acid probes and primers can be readily prepared based on the nucleic acid molecules provided in this disclosure as indicators of disease or disease progression. It is also appropriate to generate probes and primers based on fragments or portions of these nucleic acid molecules. Also appropriate are probes and primers specific for the reverse complement of these sequences, as well as probes and primers to 5' or 3' regions.

A probe comprises an isolated nucleic acid attached to a detectable label or other reporter molecule. Typical labels include radioactive isotopes, enzyme substrates, co-factors, ligands, chemiluminescent or fluorescent agents, haptens, and enzymes. Methods for labeling and guidance in the choice of labels appropriate for various purposes are discussed, *e.g.*, in Sambrook *et al.* (In *Molecular Cloning: A Laboratory Manual*, CSHL, New York, 1989) and Ausubel *et al.* (In *Current Protocols in Molecular Biology*, John Wiley & Sons, New York, 1998).

Primers are short nucleic acid molecules, for instance DNA oligonucleotides 10 nucleotides or more in length. Longer DNA oligonucleotides may be about 15, 20, 25, 30 or 50 nucleotides or more in length. Primers can be annealed to a complementary target DNA strand by nucleic acid hybridization to form a hybrid between the primer and the target DNA strand, and then the primer extended along the target DNA strand by a DNA polymerase enzyme. Primer pairs can be used for amplification of a nucleic acid sequence, *e.g.*, by the polymerase chain reaction (PCR) or other nucleic-acid amplification methods known in the art.

Methods for preparing and using nucleic acid probes and primers are described, for example, in Sambrook *et al.* (In *Molecular Cloning: A Laboratory Manual*, CSHL, New York, 1989), Ausubel *et al.* (ed.) (In *Current Protocols in Molecular Biology*, John Wiley & Sons, New York, 1998), and Innis *et al.* (*PCR Protocols, A Guide to Methods and Applications*, Academic Press, Inc., San Diego, CA, 1990). PCR primer pairs can be derived from a known sequence, for example, by using computer programs intended for that purpose such as Primer (Version 0.5, © 1991, Whitehead Institute for Biomedical Research, Cambridge, MA). One of ordinary skill in the art will appreciate that the specificity of a particular probe or primer increases with its length. Thus, for example, a primer comprising 30 consecutive nucleotides of a HRPC-related protein encoding nucleotide will anneal to a target sequence, such as another homolog of the designated HRPC-related protein, with a higher specificity than a corresponding primer of only 15 nucleotides. Thus, in order to obtain greater specificity, probes and primers can be selected that comprise at least 20, 25, 30, 35, 40, 45, 50 or more consecutive nucleotides of a HRPC-related protein-encoding nucleotide sequences.

The disclosure thus includes isolated nucleic acid molecules that comprise specified lengths of the disclosed HRPC-related nucleotide sequences. Such molecules may comprise at least 10, 15, 20, 23, 25, 30, 35, 40, 45 or 50 consecutive nucleotides of these sequences or more, and may be obtained from any region of the disclosed sequences (*e.g.*, a HRPC-related nucleic acid may be apportioned into halves or quarters based on sequence length, and isolated nucleic acid molecules may be derived from the first or second halves of the molecules, or any of the four quarters, etc.). A HRPC-related cDNA also can be divided into smaller regions, *e.g.* about eighths, sixteenths, twentieths, fiftieths and so forth, with similar effect.

Another mode of division is to select the 5' (upstream) and/or 3' (downstream) region associated with a HRPC-related gene.

Nucleic acid molecules may be selected that comprise at least 10, 15, 20, 25, 30, 35, 40, 50 or 100 or more consecutive nucleotides of any of these or other portions of a HRPC-related nucleic acid molecule, such as those disclosed herein, and associated flanking regions. Thus, representative nucleic acid molecules might comprise at least 10 consecutive nucleotides of a human coding sequence the expression of which is influenced by prostate cancer progression, such as those listed in Table 1.

Protein: A biological molecule expressed by a gene and comprised of amino acids.

Purified: The term "purified" does not require absolute purity; rather, it is intended as a relative term. Thus, for example, a purified protein preparation is one in which the protein referred to is more pure than the protein in its natural environment within a cell or within a production reaction chamber (as appropriate).

Recombinant: A recombinant nucleic acid is one that has a sequence that is not naturally occurring or has a sequence that is made by an artificial combination of two otherwise separated segments of sequence. This artificial combination can be accomplished by chemical synthesis or,

more commonly, by the artificial manipulation of isolated segments of nucleic acids, *e.g.*, by genetic engineering techniques.

Representational difference analysis: A PCR-based subtractive hybridization technique used to identify differences in the mRNA transcripts present in closely related cell lines.

Serial analysis of gene expression: The use of short diagnostic sequence tags to allow the quantitative and simultaneous analysis of a large number of transcripts in tissue, as described in Velculescu *et al.* (*Science* 270:484-487, 1995).

Sequence identity: The similarity between two nucleic acid sequences, or two amino acid sequences, is expressed in terms of the similarity between the sequences, otherwise referred to as sequence identity. Sequence identity is frequently measured in terms of percentage identity (or similarity or homology); the higher the percentage, the more similar the two sequences are. Homologs or orthologs of the HRPC-related protein, and the corresponding cDNA or gene sequence, will possess a relatively high degree of sequence identity when aligned using standard methods. This homology will be more significant when the orthologous proteins or genes or cDNAs are derived from species that are more closely related (*e.g.*, human and chimpanzee sequences), compared to species more distantly related (*e.g.*, human and *C. elegans* sequences).

Methods of alignment of sequences for comparison are well known in the art. Various programs and alignment algorithms are described in: Smith & Waterman *Adv. Appl. Math.* 2: 482, 1981; Needleman & Wunsch *J. Mol. Biol.* 48: 443, 1970; Pearson & Lipman *Proc. Natl. Acad. Sci. USA* 85: 2444, 1988; Higgins & Sharp *Gene,* 73: 237-244, 1988; Higgins & Sharp *CABIOS* 5: 151-153, 1989; Corpet *et al. Nuc. Acids Res.* 16, 10881-90, 1988; Huang *et al. Computer Appls. in the Biosciences* 8, 155-65, 1992; and Pearson *et al. Meth. Mol. Bio.* 24, 307-31, 1994. Altschul *et al.* (*J. Mol. Biol.* 215:403-410, 1990), presents a detailed consideration of sequence alignment methods and homology calculations.

The NCBI Basic Local Alignment Search Tool (BLAST) (Altschul *et al. J. Mol. Biol.* 215:403-410, 1990) is available from several sources, including the National Center for Biotechnology Information (NCBI, Bethesda, MD) and on the Internet, for use in connection with the sequence analysis programs blastp, blastn, blastx, tblastn and tblastx. By way of example, for comparisons of amino acid sequences of greater than about 30 amino acids, the Blast 2 sequences function is employed using the default BLOSUM62 matrix set to default parameters, (gap existence cost of 11, and a per residue gap cost of 1). When aligning short peptides (fewer than around 30 amino acids), the alignment should be performed using the Blast 2 sequences function, employing the PAM30 matrix set to default parameters (open gap 9, extension gap 1 penalties).

An alternative indication that two nucleic acid molecules are closely related is that the two molecules hybridize to each other under stringent conditions. Stringent conditions are sequence-dependent and are different under different environmental parameters. Generally, stringent conditions are selected to be about 5° C to 20° C lower than the thermal melting point (Tm) for the specific sequence at a defined ionic strength and pH. The T_m is the temperature (under defined ionic strength

and pH) at which 50% of the target sequence remains hybridized to a perfectly matched probe or complementary strand. Conditions for nucleic acid hybridization and calculation of stringencies can be found in Sambrook *et al.* (In *Molecular Cloning: A Laboratory Manual*, CSHL, New York, 1989) and Tijssen (*Laboratory Techniques in Biochemistry and Molecular Biology--Hybridization with Nucleic Acid Probes* Part I, Chapter 2, Elsevier, New York, 1993). Nucleic acid molecules that hybridize under stringent conditions to a specific human HRPC-related protein-encoding sequence will typically hybridize to a probe based on either an entire human HRPC-related protein-encoding sequence or selected portions of the encoding sequence under wash conditions of 2x SSC at 50° C.

Nucleic acid sequences that do not show a high degree of identity may nevertheless encode similar amino acid sequences, due to the degeneracy of the genetic code. It is understood that changes in nucleic acid sequence can be made using this degeneracy to produce multiple nucleic acid molecules that all encode substantially the same protein.

Specific binding agent: An agent that binds substantially only to a defined target. Thus a protein-specific binding agent binds substantially only the specified protein. As used herein, the term "protein [X] specific binding agent" includes anti-[X] protein antibodies (and functional fragments thereof) and other agents (such as soluble receptors) that bind substantially only to the [X] protein. In this context, [X] refers to any specific or designated protein, for instance a HRPC-related protein such as those listed in Table 1.

Anti-[X] protein antibodies may be produced using standard procedures described in a number of texts, including Harlow and Lane (*Antibodies, A Laboratory Manual*, CSHL, New York, 1988). The determination that a particular agent binds substantially only to the specified protein may readily be made by using or adapting routine procedures. One suitable *in vitro* assay makes use of the Western blotting procedure (described in many standard texts, including Harlow and Lane (*Antibodies, A Laboratory Manual*, CSHL, New York, 1988)). Western blotting may be used to determine that a given protein binding agent, such as an anti-[X] protein monoclonal antibody, binds substantially only to the [X] protein.

Shorter fragments of antibodies can also serve as specific binding agents. For instance, Fabs, Fvs, and single-chain Fvs (SCFvs) that bind to a specified protein would be specific binding agents. These antibody fragments are defined as follows: (1) Fab, the fragment which contains a monovalent antigen-binding fragment of an antibody molecule produced by digestion of whole antibody with the enzyme papain to yield an intact light chain and a portion of one heavy chain; (2) Fab', the fragment of an antibody molecule obtained by treating whole antibody with pepsin, followed by reduction, to yield an intact light chain and a portion of the heavy chain; two Fab' fragments are obtained per antibody molecule; (3) (Fab')2, the fragment of the antibody obtained by treating whole antibody with the enzyme pepsin without subsequent reduction; (4) F(ab')2, a dimer of two Fab' fragments held together by two disulfide bonds; (5) Fv, a genetically engineered fragment containing the variable region of the light chain and the variable region of the heavy chain expressed as two chains; and (6) single chain antibody ("SCA"), a genetically engineered molecule containing

the variable region of the light chain, the variable region of the heavy chain, linked by a suitable polypeptide linker as a genetically fused single chain molecule. Methods of making these fragments are routine.

Subject: Living multi-cellular vertebrate organisms, a category that includes both human and non-human mammals

Target sequence: "Target sequence" is a portion of ssDNA, dsDNA or RNA that, upon hybridization to a therapeutically effective oligonucleotide or oligonucleotide analog, results in the inhibition of expression of a specified protein, such as a HRPC-related protein. Either an antisense or a sense molecule can be used to target a portion of dsDNA, since both will interfere with the expression of that portion of the dsDNA. The antisense molecule can bind to the plus strand, and the sense molecule can bind to the minus strand. Thus, target sequences can be ssDNA, dsDNA, and RNA.

Tissue microarray ("tissue chip"): A tissue microarray is a microarray wherein the samples are samples of tissue, for instance animal tissue such as human tissue. Examples of tissue microarrays are assembled by aligning tissue cylinders (taken, for instance, from tissue blocks or biopsies) in a recipient block, such as a block of paraffin, to create a matrix of columns of sample within the block. Individual slices are cut from the surface of the block, substantially perpendicular to the axis of the cylinders, thereby yielding flat, thin arrays of tissue samples embedded in the block material. Such thin arrays are often transferred to a microscope slide or other supporting member. The construction of tissue microarrays is described in, for instance, Kononen *et al., Nature Medicine,* 4:844-847, 1998 and PCT International Patent Publication WO99/44063A2, both of which are incorporated herein by this reference.

Tissue samples contained in a tissue microarray may be any set of tissues, but often a tissue microarray has a theme so to speak, for instance containing samples from a collection of different tumors, tumors from different tissues, tumors from different stages of progression, or from different treatment regimens.

Transformed: A transformed cell is a cell into which has been introduced a nucleic acid molecule by molecular biology techniques. As used herein, the term transformation encompasses all techniques by which a nucleic acid molecule might be introduced into such a cell, including transfection with viral vectors, transformation with plasmid vectors, and introduction of naked DNA by electroporation, lipofection, and particle gun acceleration.

Tumor: A neoplasm that may be either malignant or non-malignant. "Tumors of the same tissue type" refers to primary tumors originating in a particular organ (such as breast, prostate, bladder or lung). Tumors of the same tissue type may be divided into tumor of different sub-types (an example being prostate cancer, which can be an adenocarcinoma, transitional cell, squamous cell tumor, or sarcoma).

Vector: A nucleic acid molecule as introduced into a host cell, thereby producing a transformed host cell. A vector may include nucleic acid sequences that permit it to replicate in a

host cell, such as an origin of replication. A vector may also include one or more selectable marker genes and other genetic elements known in the art.

Unless otherwise defined, all technical and scientific terms used herein have the same meaning as commonly understood by one of ordinary skill in the art to which the subject matter disclosed herein belongs. The singular terms "a", "an", and "the" include plural referents unless context clearly indicates otherwise. Similarly, the word "or" is intended to include "and" unless the context clearly indicates otherwise. Although methods and materials similar or equivalent to those described herein can be used in the practice or testing of the present disclosure, suitable methods and materials are described below. All publications, patent applications, patents, and other references mentioned herein are incorporated by reference in their entirety. In case of conflict, the present specification, including explanations of terms, will control. In addition, the materials, methods, and examples are illustrative only and not intended to be limiting.

III. Description of several specific embodiments

Provided herein are methods of diagnosing or prognosing development or progression of prostate cancer in a subject, which methods involve detecting an abnormality in at least one HRPC-related molecule of the subject (*e.g.*, an HRPC related nucleic acid molecule such as one listed in Table 1 or Table 4, or genes, cDNAs or other polynucleotide molecules comprising one of the listed sequences, or a fragment thereof, or an HRPC-related protein such as one encoded by such a nucleic acid molecule, or a fragment of such protein). In certain embodiments, abnormalities are detected in more than one HRPC-related molecule, for instance in at least 5, at least 10, 15, 25, 50, or 100 or more HRPC-related nucleic acid molecules listed in Table 1 or elsewhere herein, or encoded for by a nucleic acid molecule listed in Table 1 or elsewhere herein. In certain specific embodiments, no more than the molecules listed in Table 1 or Table 4, or corresponding to (represented by) those listed in Table 1 or Table 4, are included in such analysis. For instance, certain of the described methods employ detecting no more than 600, no more than 500, no more than 400, no more than 300, or no more than 200 of such molecules.

Also encompassed herein are arrays containing two or more HRPC-related molecules. Certain of such arrays are nucleic acid arrays, which contain at least one HRPC-related nucleic acid molecule, for instance at least one of the HRPC-related nucleic acid molecules listed in Table 1 or Table 4, or genes, cDNAs or other polynucleotide molecules comprising one of the listed sequences, or a fragment thereof. Other described arrays are protein (polypeptide) arrays, which contain at least one HRPC-related protein such as one encoded by a nucleic acid molecule listed in Table 1 or Table 4 (or genes, cDNAs or other polynucleotide molecules comprising one of the listed sequences, or a fragment thereof), or a fragment of such protein, or an antibody specific to such a protein or protein fragment. Such arrays can also contain any particular subset of the nucleic acids (or corresponding molecules) listed in Table 1 or Table 4. Certain of such arrays (as well as the methods described herein) also may include HRPC-related molecules that are not listed in Table 1 or Table 4.

Abnormalities detected by these methods for instance can be different for different HRPC-related molecules, and may include increases or decreases in the level (amount) or functional activity of such molecules, or in their localization or stability. As used herein, the term "HRPC-related molecule" includes HRPC-related nucleic acid molecules (such as DNA or RNA or cDNA) and HRPC-related proteins, though in specific embodiments the term may be specific for any one of these types of molecules. The term is not limited to those molecules listed in Table 1 or Table 4 (and molecules that correspond to those listed), but also includes other nucleic acids and/or proteins that are influenced (*e.g.*, as to level, activity, localization) by or during prostate cancer progression, including all of such molecules listed herein.

Specific encompassed embodiments include diagnostic and/or prognostic methods in which a mutation, duplication or deletion of a HRPC-related nucleic acid in cells of the individual is detected.

In certain embodiments, HRPC-related molecules that can be examined for an abnormality include molecules represented by a subset of the sequences referred to in Table 1 or Table 4, such as the more than 200 sequences represented by Image Clone ID numbers: 1475595, 1460110, 50794, 78294, 190491, 66731, 143287, 754600, 754509, 308041, 70827, 361974, 503097, 796646, 41650, 841641, 724615, 839101, 504226, 810711, 435330, 773567, 431296, 345232, 756405, 256907, 415817, 366541, 223350, 366067, 724831, 814353, 236034, 809910, 1470048, 1323448, 1456424, 453689, 135221, 340734, 180864, 768562, 179276, 44505, 293104, 243343, 66317, 812251, 245920, 265874, 770212, 784910, 839094, 712049, 669435, 841470, 782339, 297061, 429466, 300137, 487172, 343744, 795730, 268876, 742132, 755578, 502682, 510381, 140574, 135630, 278242, 742862, 1049033, 270136, 768260, 53039, 211813, 195051, 125769, 122955, 129342, 292392, 139331, 143995, 139250, 243360, 194307, 235040, 295483, 143756, 897768, 1456160, 34778, 810512, 753184, 200814, 470393, 23185, 128126, 42373, 511521, 810117, 950682, 783696, 815555, 897531, 713145, 502690, 469969, 309893, 725877, 343987, 49318, 42864, 193087, 162533, 1309620, 685801, 825740, 756708, 28469, 187147, 246304, 130280, 753587, 123980, 241985, 564621, 841507, 810703, 784772, 143306, 246722, 298417, 51582, 757222, 884783, 417424, 324891, 504791, 725877, 743230, 377048, 42627, 144797, 244955, 204735, 144747, 292749, 196109, 120375, 121981, 121715, 243403, 127409, 130053, 243291, 203514, 133130, 134495, 296552, 138601, 167076, 197323, 197637, 194906, 194985, 196125, 196303, 243784, 280122, 245235, 197856, 200604, 203400, 207448, 234469, 210548, 208940, 208434, 211951, 212098, 233399, 240138, 137396, 241097, 239835, 308231, 292312, 292391, 293421, 293306, 293785, 295044, 295590, 296102, 296602, 297110, 191572, 195132, 233274, 246546, 296562, 214331, 214043, 126230, 128245, 129616, 134312, 230613, 239711, 134537, 127646, 136984, 210610, 293457, 233299, 281125, 26184, 39093, 39884, and/or 2911545. Molecules represented by (or corresponding to) these Image Clone IDs include the nucleic acid fragments found in the respective clones (and variants thereof), complete nucleic acids (such as cDNAs, mRNAs, or genes)

encompassing such fragments, fragments and variants of these complete nucleic acid molecules, proteins encoded by such nucleic acids, and fragments and variants of such proteins.

Certain of the encompassed methods involve measuring an amount of the HRPC-related molecule in a sample (such as a serum or tissue sample) derived or taken from the subject, in which a difference (for instance, an increase or a decrease) in level of the HRPC-related molecule relative to that present in a sample derived or taken from the subject at an earlier time, is diagnostic or prognostic for development or progression of prostate cancer.

Abnormalities in HRPC-related nucleic acid molecules can be detected using, for instance, *in vitro* nucleic acid amplification and/or nucleic acid hybridization. The results of such detection methods can be quantified, for instance by determining the amount of hybridization or the amount of amplification.

Abnormalities in HRPC-related proteins can be detected using, for instance, a HRPC protein-specific binding agent, which in some instances will be detectably labeled. In certain embodiments, therefore, detecting an abnormality includes contacting a sample from the subject with a HRPC protein-specific binding agent; and detecting whether the binding agent is bound by the sample and thereby measuring the levels of the HRPC-related protein present in the sample, in which a difference in the level of HRPC-related protein in the sample, relative to the level of HRPC-related protein found an analogous sample from a subject not having the disease or disorder, or a standard HRPC-related protein level in analogous samples from a subject not having the disease or disorder or not having a predisposition for developing the disease or disorder, is an abnormality in that HRPC-related molecule.

In other embodiments, detecting the abnormality involves determining whether a HRPC-related gene expression profile from the subject indicates development or progression of prostate cancer, for instance by comparing the HRPC-related gene expression profile from the subject to at least one control gene expression fingerprint or profile for a specific stage of prostate cancer. In specific examples of such methods, at least one control gene expression profile is a fingerprint for a normal prostate tissue, a primary prostate cancer tissue, a prostate cancer tissue responding to androgen ablation therapy, or a hormone refractory prostate cancer tissue. Examples of such profiles (also referred to herein as fingerprints) can be in an array format, such as a nucleotide (*e.g.*, polynucleotide) or protein array or microarray, or generated from such an array.

Specific embodiments of methods for detecting an abnormality in at least one HRPC-related molecule use the arrays disclosed herein. Such arrays are nucleotide (*e.g.*, polynucleotide) or protein (*e.g.*, peptide, polypeptide, or antibody) arrays. In such methods, an array may be contacted with polynucleotides or polypeptides (respectively) from (or derived from) a sample from a subject. The amount and/or position of binding of the subject's polynucleotides or polypeptides then can be determined, for instance to produce a gene expression profile for that subject. Such gene expression profile can be compared to another gene expression profile, for instance a control gene expression profile from a subject having a known prostate-related condition. Optionally, the subject's gene

expression profile (also known as a gene expression fingerprint) can be correlated with one or more appropriate treatments. Similarly, protein arrays can give rise to protein expression profiles. Both protein and gene expression profiles can more generally be referred to as expression profiles.

Other embodiments are methods that involve providing nucleic acids from the subject; amplifying the nucleic acids to form nucleic acid amplification products; contacting the nucleic acid amplification products with an oligonucleotide probe that will hybridize under stringent conditions with a nucleic acid encoding a HRPC-related protein; detecting the nucleic acid amplification products which hybridize with the probe; and quantifying the amount of the nucleic acid amplification products that hybridize with the probe. The sequence of such oligonucleotide probes may be selected to bind specifically to a nucleic acid molecule listed in Table 1 or Table 4, or a nucleic acid molecule represented by those listed in Table 1 or Table 4. In some embodiments, the probes are attached to a solid surface, such as an array. Likewise, the primers may be selected to amplify a nucleic acid molecule listed in Table 1 or Table 4, or represented by those listed in Table 1 or Table 4. In specific examples of such methods, the primers are selected to amplify a nucleic acid product encoding cartilage glycoprotein-39 (CHI3L1), S-100P PROTEIN (S100P), CX3C chemokine/fractalkine (SCYD1), adenylate kinase 1 (AK1), forkhead transcription factor HFH-4 (HFH-4) (FKHL13), UDP glucuronosyltransferase precursor (UGT2B15), Pleiotrophin (heparin binding growth factor 8) (PTN), heat shock 27kD protein 2/Alpha-B-crystallin (HSP27), Proteasome (prosome, macropain) subunit, beta type, 5 (PSMB5), Inhibitor of NFKB (NFKBIA), interferon-induced 17 kD protein (ISG15), MAP kinase activated protein kinase 2 (MAPKAPK2), signal transduction protein (SH3 containing) (EFS2), hkf-1 Zinc finger protein (ZFP103), chromosome condensation 1 (CHC1), CDP-diacylglycerol synthase (CDS1), gap junction protein, alpha 1, 43 kD (connexin 43) (GJA1), cyclin D1 (CCND1), Inhibitor of DNA binding 3, helix-loop-helix protein (ID3), H1 histone family, member2 (H1F2), Cytochrome B561 (CYB561), Cathepsin H (CTSH), calcineurin alpha (PPP3CA), 54 kDa progesterone receptor-associated immunophilin (FKBP5), translocation protein 1 (TLOC1), Clusterin (complement lysis inhibitor; testosterone-repressed prostate message 2; apolipoprotein J) (CLU), Pulmonary surfactant-associated protein A (SFTPA1), protease inhibitor 12 (PI12; neuroserpin) (PI12), Thrombospondin 1 (THBS1), Ribophorin I (RPN1), A disintegrin-like and metalloprotease (reprolysin type) with thrombospondin type 1 motif, 1 (ADAMTS1), Collagen, type IV, alpha 5 (Alport syndrome) (COL4A5), LIM domain only 4 / breast tumor autoantigen (LMO4), bumetanide-sensitive Na-K-Cl cotransporter (NKCC1) (SLC12A2), Fibronectin (FN1), Crystallin Mu (CRYM) or "upregulated by 1,25-dihydroxyvitamine D-3 (VDUP1).

Also encompassed are methods of prostate cancer therapy, in which an abnormality in at least one HRPC-related molecule of a subject is detected using a method described herein, and; if such abnormality is identified, a treatment is selected to prevent or reduce hormone-refractory prostate cancer or to delay the onset of hormone-refractory prostate cancer. The subject then can be treated in accordance with this selection. In some examples, the treatment selected in specific and

tailored for the subject, based on the analysis of that subject's profile for one or more HRPC-related molecules.

A further embodiment is a method of modifying a level of expression or function of a HRPC-related protein in a subject. Such methods may involve expressing in the subject a recombinant genetic construct that includes a promoter operably linked to a nucleic acid molecule, and wherein expression of the nucleic acid molecule changes expression of the HRPC-related protein. The nucleic acid molecule may, for instance, include at least 10 consecutive nucleotides of a HRPC-related nucleic acid sequence. In specific examples of such methods, the nucleic acid molecule is in antisense orientation relative to the promoter; in other examples, the nucleic acid molecule is in sense orientation relative to the promoter.

Other embodiments are kits for measuring the level or function of one or more HRPC-related molecules, which kits may include a binding molecule that selectively binds to the HRPC-related molecule that is the target of the kit. In some examples of such kits where the HRPC-related molecule level is a HRPC-related protein level, the binding molecule provided in the kit may be an antibody or antibody fragment that selectively binds to the target HRPC-related protein. In other examples of such kits where the HRPC-related molecule level is a HRPC-related nucleic acid molecule level, the binding molecule provided in the kit may be an oligonucleotide capable of hybridizing to the HRPC-related nucleic acid molecule.

Further embodiments are methods of screening for a compound useful in treating, reducing, or preventing prostate cancer or development or progression of prostate cancer. Such methods involve determining if application of a test compound alters a HRPC-related gene expression profile so that the profile more closely resembles a prostate-linked profile than it did prior to such treatment, and selecting a compound that so alters the HRPC-related gene expression profile. In specific examples of such methods, the test compound is applied to a test cell. In some of such methods, the profile is determined or measured in an array format.

Also encompassed are compounds selected using the methods described herein, which are useful in treating, reducing, or preventing prostate cancer or development or progression of prostate cancer.

Examples of disclosed methods involve contacting test cells with a test compound, then measuring at least one HRPC-related molecule level and/or activity in the test cells. In such methods, a difference in HRPC-related molecule (*e.g.*, a HRPC-related nucleic acid molecule listed in Table 1, or a molecule encoded for by a nucleic acid molecule listed in Table 1) level and/or activity in the test cells, relative to the analogous HRPC-related molecule level and/or activity found in analogous cells not contacted with the test compound, indicates that the test compound is useful in treating, reducing, or preventing prostate cancer or development or progression of prostate cancer. Measuring the HRPC-related molecule(s) level and/or activity may include creating a HRPC-related gene expression profile for the test cell after contacting the cell with the test compound, and comparing the test cell HRPC-related gene expression profile to at least one control gene expression profile for a specific

stage of prostate cancer. Representative control gene expression profile can include a profile for a normal prostate tissue, a primary prostate cancer tissue, a prostate cancer tissue responding to androgen ablation therapy, and/or a hormone refractory prostate cancer tissue.

Also disclosed herein are use of identified target HRPC-related molecules for the development of antibodies, including therapeutic antibodies that affect an HRPC-related pathway. It is also envisioned that the disclosed HRPC-related molecules can be used as vaccines, for instance as "cancer vaccines" to elicit an immune response from a subject that renders the subject more resistant to developing or progressing through a stage of prostate cancer.

IV. Temporal gene expression changes during prostate cancer hormonal therapy

The present disclosure concerns gene expression profiling of the regression and progression of the CWR22 human prostate cancer xenografts using cDNA microarrays. Highly quantitative two-color fluorescent microarray technology was used to examine differences in gene expression between primary and recurrent CWR22 prostate cancer xenografts. cDNA microarrays containing 6605 gene fragments were constructed and applied to analyze the mRNA expression profile of thirteen CWR22 xenografts at different stages in the time course of treatment and progression. New statistical tools (including template-based gene clustering) were then used to mine this data (*e.g.*, to differentiate temporal gene expression profiles associated with *in vivo* therapy response from the noise in the data), enabling a comprehensive genomics-based analysis of the temporal mRNA expression profiles during the entire course of CWR22 treatment and progression.

For instance, a large set of genes and other encoding sequences (*e.g.*, expressed sequence tags, ESTs) have been identified (Table 1), the expression of which varies during prostate cancer progression. The Image Clone ID numbers of these sequences are: 843249, 796904, 399604, 788205, 260303, 287745, 796542, 243653, 486787, 134495, 796680, 448386, 298417, 124578, 141562, 713145, 815284, 51408, 22012, 344589, 839101, 511091, 365515, 144042, 153411, 853809, 884783, 75254, 109314, 293925, 949938, 132835, 773567, 415529, 809910, 142326, 139660, 435855, 243816, 489839, 130895, 258120, 39920, 343867, 142139, 1472735, 503083, 796147, 813179, 741891, 461727, 756968, 814636, 897667, 297392, 322723, 66322, 295729, 265645, 143756, 195340, 78294, 296880, 713922, 203772, 383188, 431296, 842939, 347434, 308041, 202535, 66582, 795730, 292770, 788107, 128143, 214162, 298231, 453183, 502518, 755054, 415978, 630013, 813678, 236034, 257162, 813841, 770074, 266146, 377692, 755952, 297063, 840511, 415084, 841695, 183462, 731051, 80221, 758222, 71116, 137139, 136557, 205239, 509731, 208718, 840460, 111750, 711918, 809998, 784296, 813266, 257422, 82734, 811161, 322160, 844725, 276091, 177772, 461804, 898258, 248412, 344139, 796689, 223350, 78353, 502832, 250654, 214990, 813611, 140806, 1469234, 781362, 813698, 377731, 79782, 768644, 772425, 150702, 246430, 210494, 120681, 132140, 431501, 366966, 35105, 201393, 137096, 453005, 131050, 897485, 781233, 768246, 897906, 246765, 1412481, 111150, 124071, 42070, 1455976, 66982, 362483, 382195, 196992, 564621, 814409, 504763, 143519, 82903, 234736, 812246, 125685, 436155, 898219, 773637, 269878, 427812, 868368, 81475, 784910, 859359, 809353, 753069,

377320, 1472775, 130826, 46284, 586685, 811162, 50680, 840990, 1461664, 796613, 68977, 796198, 80338, 770394, 138936, 784593, 768299, 232772, 32493, 432210, 126858, 245936, 1161775, 840942, 361974, 51406, 41650, 77728, 278570, 271670, 26249, 810331, 811088, 627306, 179890, 121454, 289447, 810104, 774471, 377252, 949928, 294881, 292364, 142788, 246619, 345935, 854899, 770059, 49630, 773301, 781139, 854338, 785967, 770388, 810960, 383175, 128460, 279970, 139217, 283023, 813823, 242062, 461327, 756595, 826142, 841008, 134229, 1323448, 300137, 843098, 51582, 128785, 839991, 783998, 130276, 214006, 840687, 66731, 272327, 197054, 814240, 121792, 809456, 145001, 685371, 345626, 136954, 143887, 120964, 842784, 1374571, 152453, 842836, 233583, 346552, 51293, 360885, 815774, 852829, 35077, 488956, 31143, 741067, 725877, 345858, 35828, 34849, 431397, 203469, 713782, 296754, 768370, 246546, 66686, 547058, 144924, 767475, 591281, 1474337, 823691, 812955, 1416782, 232670, 183602, 1343468, 289818, 486186, 784772, 491565, 897781, 1472150, 745343, 788256, 1461138, 742798, 214965, 842968, 490778, 842863, 415102, 825606, 196303, 490995, 564803, 292933, 841507, 415089, 785707, 1492304, 884867, 774446, 781047, 811046, 727526, 126650, 233274, 838802, 232837, 45544, 280934, 294487, 795936, 141818, 131316, 700792, 969748, 340558, 34778, 28469, 795498, 293569, 47853, 200402, 47853, 852520, 825295, 26184, 1456160, 491001, 757489, 321661, 592359, 51448, 725284, 46182, 324891, 811015, 36393, 207288, 744417, 813648, 745138, 612274, 785616, 214614, 869538, 71101, 204257, 853368, 769921, 320509, 249603, 207358, 435076, 242578, 139705, 299360, 753457, 810899, 34355, 291057, 43550, 826211, 789147, 295483, 797016, 309288, 44975, 416833, 897567, 809588, 772304, 323404, 809603, 295986, 725188, 744047, 783697, 814615, 814701, 898286, 950690, 66564, 454339, 204214, 796646, 129865, 756401, 66406, 451907, 827144, 25584, 365641, 840894, 782679, 711768, 234237, 416833, 884718, 788185, 453107, 204148, 509887, 289551, 740554, 211747, 66728, 789204, 362926, 878676, 611150, 50506, 162775, 743230, 626716, 47833, 796278, 128243, 80946, 149013, 531319, 950482, 950473, 789182, 856427, 725454, 951142, 878130, 49352, 322914, 1472643, 293727, 273546, 772220, 53316, 42059, 810854, 768260, 626531, 471598, 44537, 769603, 376785, 760344, 840364, 856489, 490772, 46171, 855487, 281003, 509495, 43231, 487348, 898062, 795738, 24145, 40017, 429182, 825677, 755239, 971367, 129146, 825312, 384081, 26578, 814287, 787938, 857264, 813675, 134719, 626206, 684655, 29063, 433666, 42096, 325641, 246120, 80410, 1160558, 45233, 214884, 824906, 814117, 810057, 1455641, 545403, 773383, 840702, 810552, 739511, 145503, 135449, 724387, 283315, 897177, 866874, 502669, 324618, 897774, 73381, 41452, 321389, 949971, 785778, 50359, 813280, 308682, 531957, 486175, 40026, 28823, 487425, 42880, 416316, 32875, 753862, 795543, 263727, 824568, 366156, 1492412, 840567, 51666, 72050, 47647, 809535, 810725, 842825, 767817, 80500, 856454, 811150, 470930, 242698, 83279, 884993, 211275, 741958, 433474, 196650, 782439, 843121, 28410, 378502, 214906, 43241, 47542, 109863, 814765, 384015, 489823, 83011, 134544, 711552, 195051, 268727, 742132, 108265, 280837, 770837, 241988, 66555, 208413, 399532, 291880, 814731, 42313, 433350, 415145, and 2911545. These Image ID Clones can be obtained from Research Genetics (2130 Memorial Parkway, Huntsville, AL 35801, US or Canada: 1-

800-533-4363). The sequences of these Image ID Clones are hereby expressly incorporated by reference, and are clearly identified based on their multiple identification listings given in Table 1.

Up to 231 (3.5%) of the 6605 coding sequences assayed were differentially expressed between primary and recurrent xenografts. Using data from the direct hybridization of mRNA from a pool of four primary CWR22 xenografts against a pool of four CWR22R recurrent xenografts, the data was filtered for an intensity of at least greater than the mean in the maximum between the cy5 and cy3 signals. The mean intensity was 3058, which was used as a cut-off to ensure that the data was of high quality, however other intensities may be used. This intensity cut-off resulted in 2276 genes with sufficient expression. Based on the 99.9% confidence level, 231 of these 2276 coding sequences were considered differentially expressed though prostate progression. These include a set of 90 sequences that display reduced cDNA production after hormone therapy with an increase (are up-regulated) during tumor recurrence (Image ID Clones: 1475595, 1460110, 50794, 78294, 190491, 66731, 143287, 754600, 754509, 308041, 70827, 361974, 503097, 796646, 41650, 841641, 724615, 839101, 504226, 810711, 435330, 773567, 431296, 345232, 756405, 256907, 415817, 366541, 223350, 366067, 724831, 814353, 236034, 809910, 1470048, 1323448, 1456424, 453689, 135221, 340734, 180864, 768562, 179276, 44505, 293104, 243343, 66317, 812251, 245920, 265874, 770212, 784910, 839094, 712049, 669435, 841470, 782339, 297061, 429466, 300137, 487172, 343744, 795730, 268876, 742132, 755578, 502682, 510381, 140574, 135630, 278242, 742862, 1049033, 270136, 768260, 53039, 211813, 195051, 125769, 122955, 129342, 292392, 139331, 143995, 139250, 243360, 194307, 235040, 295483, and 143756), and a set of 131 sequences that display increased cDNA production after hormone therapy with a decrease (are down-regulated) during tumor recurrence (Image ID Clones: 897768, 1456160, 34778, 810512, 753184, 200814, 470393, 23185, 128126, 42373, 511521, 810117, 950682, 783696, 815555, 897531, 713145, 502690, 469969, 309893, 725877, 343987, 49318, 42864, 193087, 162533, 1309620, 685801, 825740, 756708, 28469, 187147, 246304, 130280, 753587, 123980, 241985, 564621, 841507, 810703, 784772, 143306, 246722, 298417, 51582, 757222, 884783, 417424, 324891, 504791, 725877, 743230, 377048, 42627, 144797, 244955, 204735, 144747, 292749, 196109, 120375, 121981, 121715, 243403, 127409, 130053, 243291, 203514, 133130, 134495, 296552, 138601, 167076, 197323, 197637, 194906, 194985, 196125, 196303, 243784, 280122, 245235, 197856, 200604, 203400, 207448, 234469, 210548, 208940, 208434, 211951, 212098, 233399, 240138, 137396, 241097, 239835, 308231, 292312, 292391, 293421, 293306, 293785, 295044, 295590, 296102, 296602, 297110, 191572, 195132, 233274, 246546, 296562, 214331, 214043, 126230, 128245, 129616, 134312, 230613, 239711, 134537, 127646, 136984, 210610, 293457, 233299, 281125, 26184, 39093, and 39884). Other confidence levels could be used to select HRPC-related molecules, such as 98%, 95%, 90%, 85%, and so forth. Higher confidence levels, such as 99.99%, could also be used. Molecules identified as being linked to prostate cancer (referred to generally herein as HRPC-related molecules) using the methods described herein can be arranged on arrays for use in diagnostic and prognostic

methods. Specific arrays are contemplated that are constructed using molecules identified at such different confidence levels.

In particular, the techniques disclosed herein have uncovered many genes not previously associated with prostate cancer progression, and particularly not previously associated with HRPC. These newly correlated genes include those represented by the following Image ID Clones: 781047, 785778, 842968, 769921, 898286, 204214, 814701, 435076, 531319, 415089, 898062, 453107, 785707, 795936, 700792, 34778, 46182, 769921, 783697, 451907, 711768, 416833, 810711, 789204, 789182, 725454, 951142, 49352, 273546, 46717, 855487, 41117, 26578, 684655, 45233, 814117, 810552, 739511, 283315, and 897774.

Of the 59 androgen-dependent sequences whose expression decreased most after castration (labeled "Decreasing" in Table 2), 58 (98.3%) displayed restored transcript levels in the recurrent tumors, indicating re-activation of androgen-dependent genes in the absence of a ligand.

Tissue microarrays consisting of 50 xenografts and 440 clinical specimens from all stages of prostate cancer progression were utilized to validate potential drug target genes using mRNA *in situ* hybridization and protein immunohistochemistry. Measured by cDNA microarrays, S100P (encoding a calcium-binding protein) was among the most highly overexpressed genes in the CWR22R recurrent tumors; this gene was also highly expressed in the majority of hormone-refractory clinical prostate cancers, but rarely (<10%) in benign prostate lesions.

The temporal gene expression changes identified herein facilitate identification of candidate drugs for hormone-refractory prostate cancer. FKBP5 for example was identified and its utility as a therapeutic target was validated using tissue microarray analysis (see Example 3). Based on such leads, Rapamycin, MS-275, and TSA were tested for their effectiveness in influencing prostate cancer cell growth. These drugs target some of the candidate genes described herein. As described in Example 4, the inventors found that these drugs inhibit CWR22R prostate cancer cell growth *in vitro*. Thus, incorporating cDNA microarray technologies for genomics-based discovery of therapy response genes, with high throughput tissue microarray analysis, provides a new paradigm to identify, prioritize, and validate novel diagnostic and drug targets, as herein described for hormone-refractory prostate cancer.

The identified HRPC-related genes represent putative mediators of hormone therapy response and resistance, and as such are candidate targets for the development of novel therapeutics to maintain prostate cancer in regression following hormone ablation therapy. The utility of these genes as candidate drug targets and biomarkers is demonstrated herein by first using tissue microarrays for high throughput translation to clinical samples, and then selecting drugs that might target these genes. Analysis of cDNA microarray data with template based gene clustering and high throughput translation using tissue microarrays introduces a new, generally applicable paradigm for applying functional genomics to identify genetic programs that mediate a responses to a variety of *in vivo* therapies.

It is contemplated that certain of the HRPC-related genes identified herein encode or correspond to soluble proteins, while other encode or correspond to membrane associated or membrane integral proteins, some of which are exposed at least to a certain extent on the exterior of a cell in which they are expressed. In some embodiments, those HRPC-related molecules that are expressed at or on the surface of a cell are selected as therapeutic targets, for instance for targeting with an antibody-based therapy, which is facilitated by the access of the HRPC-related molecule to the extracellular matrix. These HRPC-related molecules may be described as being "drug accessible."

The disclosure is further illustrated by the following non-limiting Examples.

EXAMPLE 1

Identification of Genes with Altered Expression in Hormone Refractory Prostate Cancer

This example provides a description of how the disclosed HRPC-related nucleic acid molecules were identified. These HRPC-related nucleic acid molecules show differences in expression during prostate cancer development, and particularly during hormone ablation therapy and subsequent progression to a hormone-refractory condition.

Methods and Material:

Xenografts and Cell Line: CWR22 is a serially transplantable, prostate cancer xenograft that was derived from a Gleason score 9 primary human prostate cancer with osseous metastasis (Wainstein *et al.*, *Cancer Res.* 54:6049-6052, 1994). CWR22 is highly responsive to androgen deprivation, with marked tumor regression after castration (Cheng *et al.*, *J. Natl. Cancer Inst.* 88:607-611, 1996; Nagabhushan *et al.*, *Cancer Res.* 56:3042-3046, 1996; Myers *et al.*, *J Urol.* 161:945-949, 1999). About half of the treated animals develop recurrent tumors (CWR22R) over a time frame of from a few weeks to several months. CWR22R is not dependent on androgen and is able to grow in castrated animals.

Thirteen fresh-frozen human prostate xenograft tissues were recovered from mice at different stages of hormonal therapy (four primary untreated CWR22, CWR22 after 0.5 days, 2 days, 4 days, 8 days and 16 days after castration, and four independent hormone-refractory CWR22R strains). LNCap (ATCC) and CWR22R (established from recurrent CWR22R xenografts) cell lines were cultured in RPMI1640 (BibcoBRL) with 10% Fetal Bovine Serum (GibcoBRL) at 37 °C and 5% CO_2. The tumors were flash frozen and stored at -70 °C. RNA was extracted by crushing the tumors in liquid nitrogen and used directly for mRNA isolation with the FastTrack 2.0 Kit (Invitrogen Corp., Carlsbad, CA).

cDNA Microarrays: The cDNA microarrays consisted of 6605 elements representing different (non-redundant) genes. PCR products from sequence-verified clones (Research Genetics, Huntsville AB) were prepared and printed at high density onto glass slides according to previously described protocols (Mousses *et al.*, "Gene Expression Analysis by cDNA Microarrays," in

Differential Gene Expression: A practical approach, Livesey and Hunt (eds.), Oxford University Press, 2000).

Labeled cDNA was made with 4-16 μg of mRNA in an oligo(dT)-primed polymerization using SuperScript II reverse transcriptase (LifeTechnologies, Rockville , MD) in the presence of either Cy3 or Cy5 labeled dUTP (Amersham Pharmacia, Piscataway, NJ) as described (Mousses *et al.*, "Gene Expression Analysis by cDNA Microarrays," in *Differential Gene Expression: A practical approach*, Livesey and Hunt (eds.), Oxford University Press, 2000). The standard reference cDNA (Cy5 labeled LNCap cDNA) and the Cy3 labeled test cDNA from a xenograft mRNA were simultaneously hybridized to the microarray according to the protocol described previously (Mousses *et al.*, "Gene Expression Analysis by cDNA Microarrays," in *Differential Gene Expression: A practical approach*, Livesey and Hunt (eds.), Oxford University Press, 2000).

Imaging and Image Analysis: Fluorescence intensities at the immobilized targets were measured by using a custom-designed laser con-focal microscope scanner, with intensity data integrated over 15-micron square pixels and recorded at 16 bits. Image analysis was performed by using DEARRAY software. Details of the fabrication of the microarray slides, and image generation and analysis are available on the Internet at the NHGRI Microarray Website, and software is currently and freely available for Sybase/UNIX and is in the process of being ported to Oracle/UNIX. Detailed information about the program itself can be found on the ArrayDB Web site at the NHGRI. A complete description of the gene clustering used is also described at the NHGRI Microarray Website.

In brief, clustering analysis is a powerful tool that partitions biological samples or genes into well-separated and homogeneous groups based on their statistical behaviors. The main objective of clustering analysis is to find out the similarities between experiments or between each genes, given their expression ratios across all genes or samples, respectively, and then group the similar samples or genes together for the convenience of understanding and visualization. The clustering methods have been heavily studied for many years and widely applied in many areas.

Hierarchical Clustering methods: Assume there are m expression experiments containing n genes in each every experiment. After performing microarray image analysis and data integration, a $m \times n$ matrix of gene expression ratios is obtained, where each column of ratios represents the result from one expression experiment comparing the test sample to a common reference sample of choice. To simplify the discussion, the algorithm is considered only in terms of the sample clustering.

To achieve the objective of clustering, all pair-wise similarities between samples are evaluated first, and then an "average linkage algorithm" is employed to group similar samples. Typically, a Pearson correlation coefficient or Euclidean distance is used to quantify the similarity. Under certain normalization condition, these two similarity measurements are equivalent. After evaluating similarities from all pairs of samples, a distance matrix can be constructed as shown below (Table 3a). The hierarchical algorithm proceeds as follows: First a pair of experiments with shortest distance or most similarity in gene expression pattern are identified (Exp1 and Exp2 in Table 3a). A

"composite experiment" is then constructed by averaging (thus the term average-linkage algorithm) all gene expression ratios (log-transformed) from two experiments. This is referred to as Exp1-2 in the example. All distances from this composite experiment to all other experiments are then examined, and used to construct a smaller matrix, as shown in Table 3b. This procedure is repeated until the distance matrix is reduced to single element.

TABLE 3a

	Exp 1	Exp 2	Exp 3
Exp 1	0		
Exp 2	0.1	0	
Exp 3	0.7	0.4	0

TABLE 3b

	Exp 1-2	Exp 3
Exp 1-2	0	
Exp 3	0.55	0

The graphical visualization of the hierarchical algorithm is illustrated by a dendrogram, where each merger is represented by a binary tree, and the length of each branch is indicative of the distance between two samples, such as those given in Table 3a and 3b.

Template Based Gene Clustering algorithm:

To fully exploit the characteristics of temporal response of gene expression to a given treatment, either an instantaneous stimulation or a continuously increasing/decreasing excitation, a sequence of pre-ordered templates which reflect all possible gene expression responses for a given stimulation was employed. The objective of the template-based algorithm is, given the *k*th gene's temporal expression profile, to evaluate the similarities to all of ordered templates, and then based on the similarities of all templates, to produce a template index and a best similarity measure based on the Pearson correlation coefficient. Let the temporal expression profile for *k*th gene is $g_k(t_n)$ ($\log_{10}$-transformed expression ratios), and the *i*th response template to be $T_i(t_n)$, $n = 1, \ldots, N$. The similarity between the *k*th gene expression profile and *i*th template is defined by,

$$\rho_{k,i} = \frac{\sum_n \left(g_k(t_n) - \mu_{g_k}\right)\left(T_i(t_n) - \mu_{T_i}\right)}{N\sigma_{g_k}\sigma_{T_i}}$$

where μ and σ are means and standard deviations, respectively, for *k*th gene expression profile and *i*th template pattern across *N* time points. For a given gene *k*, the best similarity ρ_k from all templates is

$$\rho_k = \max_i \{\rho_{k,i}\}$$

and let the I^* to be the template that satisfies $\rho_{k,I^*} = \rho_k$, the template index is α_k for gene *k*,

$$\alpha_k = \frac{\sum_{i=I^*-2}^{I^*+2} i\rho_{k,i}}{\sum_{i=I^*-2}^{I^*+2} \rho_{k,i}}.$$

Usually, α_k indexes to somewhere near the best-fit template index I^*, but adjusted according to the similarity of its neighboring templates given the pre-defined order. The predicted fold-change of gene expression profile is also defined based on the best-fitted template I^* to be $F_k = 10^{bk}$ where b_k is the slope of the regression line.

Typically, the aforementioned template-based algorithm provides three parameters for each gene for a given order of template sequence. They are α_k, ρ_k, I^*, and F_k for template index, best Pearson's similarity measure, the best fit template, and the predicted fold-change derived from the best fitted template, respectively. Given the characteristics of these parameters, we can easily perform following data analysis: 1) sorting the α_k to order the gene expression profiles; 2) eliminating genes with small ρ_k or small F_k since their temporal expression profiles do not resemble close enough to any of the templates, and/or simply do not respond to the stimulation; and 3) studying the template given by I^* for the property of gene functions.

A data-mining tool was developed to investigate the experimental model of hormone tumor therapy *in vivo*. This model consists of independent clones of the same tumor undergoing different fates. A method for comparing the cDNA microarray data across the independent clones that undergo different fates was therefore needed to identify genes with temporal expression profiles associated with the response / phenotype of therapy *in vivo*. Filtering variables at different stringencies was used to mine the data to identify the genes that change most significantly in a manner that best reflects the temporal nature of the phenotypic changes observed during androgen ablation therapy.

Three criteria were used to mine the data to find genes that are associated with the phenotype: Variables assigned to each profile to facilitate data mining and clustering: the maximum correlation coefficient; the cluster location; and the fold change in ratio.

The link to phenotype was accomplished by filtering the "maximum correlation coefficient" to templates that best describe the temporal profile of the phenotype. This also allows noise to be filtered out.

Clustering was accomplished by sorting the "cluster location". This organized the genes/templates so genes with similar profiles are clustered together. The cluster position was calculated by the weighted average of the three template positions that had the best correlation. For example, two different gene-expression profiles may have a maximum correlation coefficient for template number 5, but have different cluster locations such as 4.6 and 5.4, allowing for a continuum of locations between the templates.

The data can be mined further by filtering the data for "Fold Change" and "Fold Change to Recurrence." In this way, the amplitude of the change can be used to increase the stringency of the filter and identify genes that change most significantly. By filtering for Fold Change to Recurrence, genes were isolated that not only have a kinetic that fits the regression phenotype, but that are also restored in recurrent tumors.

Northern Analysis Xenograft and cell line mRNA (4 μg) was subjected to electrophoresis in a formamide containing agarose gel and blotted onto a nylon membrane (Hybond-N from Amersham) and probed according to the manufacturers protocol. An 342 bp S100P-specific DNA probe was PCR amplified with S100P specific primers (SEQ ID NO: 1, also referred to as S100PF, and SEQ ID NO: 2, also referred to as S100PR) from cDNA made with Superscript II reverse transcriptases using the

manufacturers protocol (LifeTechnologies, Rockville , MD). The probe was radiolabeled with ^{32}P (NEN) using the Oligolabelling Kit (Pharmacia Biotechemicals Piscataway, NJ) according to the manufacturers specifications.

IHC and mRNA in situ using Tissue Microarray The prostate tissue microarray was constructed from paraffin embedded tumor tissue and benign control specimens. The tissue microarray permits analysis of up to 600 specimens simultaneously, greatly facilitating high throughput analysis of molecular markers in cancer tissue. The prostate tissue specimens were obtained from the Institutes of Pathology, University of Basal (Switzerland) and Tampere University Hospital (Finland). One pathologist (L. Bubendorf) reviewed all original tissue sections. The tissue microarray representing prostate neoplasm progression was constructed with 0.6 mm tissue cores and precisely arranged on a standard glass slide as described by Kononen *et al.* (*Nat. Med.* 4:844-847, 1998). The microarray tissue samples represented 45 Benign Prostate Hypertrophy (BPH), 60 prostate intraepithelial neoplasia (PIN), 264 primary tumors, 134 hormone refractory tumors, and 41 metastatic tumors. Additionally, the tissue microarray contained 28 xenograft CRW tumor specimens and several other xenografts. Protocols for preparing prostate tissue microarrays are provided for instance in Bubendorf *et al.*, *J Natl Cancer Inst* 91:1758-1764, 1999 and Bubendorf *et al.*, *Cancer Res.* 59:803-806, 1999.

The tissue microarray facilitates simultaneous application of molecular diagnostic techniques, such as immunohistochemistry. Antigen retrieval was performed by treatment in a pressure cooker for 30 minutes. Standard indirect immunoperoxidase procedures were used for immunohistochemistry (Envision Plus, DAKO). A monoclonal mouse antibody (1:1000, Transduction Laboratories, Lexington, KY) was used for detection of S100P. The reactions were visualized with diaminobenzidine as a chromagen. The nuclear and cytoplasm staining intensity were classified into three groups (negative, weak and strong staining) in duplicate by two pathologists (Hostetter, G. and Ferhle, W.). Other antibodies used for IHC on tissue arrays included Ki67, S100P, FKBP5, PCNA, PSA, AR.

Representative primers used for S100P mRNA *in situ* hybridization were as follows:
AntiS100P-A: C ATGCCCATGGCTGTCTCTAGTTCCGTCATGGTGCTAG (SEQ ID NO: 3);
AntiS100P-B: CGTGCTGCCCTCGCTGCCCGAATATCGGGAAAAGACGTCTATGAT (SEQ ID NO: 4);
AntiS100P-C: TTATCCACGGCATCCTTGTCTTTTCCACTCTGCAGG (SEQ ID NO: 5);
AntiS100P-D: TCCACCTGGGCATCTCCATTGGCGTCCAGGTCCTTGAGCA (SEQ ID NO: 6);
AntiS100P-E: AGACGTGATTGCAGCCACGAACACGATGAACTCACTGAAG (SEQ ID NO:7);
AntiS100P-F: CATTTGAGTCCTGCCTTCTCAAAGTACTTGTGACAGGC (SEQ ID NO: 8);
AntiS100P-G: GGGACCATGGCTCTGCAGGAATCTGTGACATCTCCAGGGC (SEQ ID NO: 9);
and AntiS100P-H: GCTCAGCCTAGGGGAATAATTGCCAACAAACACTTTTGGGAAGCC (SEQ ID NO: 10).

The scatter plot shown in FIG 2 was generated for one primary and one recurrent tumor, to demonstrate the correlation between the samples (low variance as indicated by lack of scatter) and to highlight the most differentially expressed genes.

Results

Therapy-associated phenotype: The CWR22 xenografts were serially passed in nude mice. Tumor material was harvested from mice at (1) primary, (2) androgen withdrawal therapy induced regression, and (3) recurrent stages. Ki67 staining of these tissues indicated that the number of cells that were proliferating (Ki67 staining positive) decreased gradually, reaching a minimum at day eight, where almost all cells were negative for Ki67 staining (FIG 1D). All the recurrent tumors had Ki67 staining that was higher than the primary tumors and approached 100% of the cells. Similar results were seen with PCNA (Myers *et al.*, *J Urol.* 161:945-949, 1999), indicating that proliferation was shut off in the recurrent tumors but the entire tumor did not respond fully to the therapy until four to eight days. Androgen receptor (AR) immunohistochemical (IHC) staining (as described above), and northern analysis (as described herein) showed only a small increase in AR mRNA and protein after castration. In general, AR protein levels were about two-fold higher in the recurrent tumors.

cDNA Microarray Analysis with Template Based Gene Clustering

For these analyses, cDNA microarrays containing sequence-verified clones that represent 6605 unique genes were constructed. Fluorescence intensity ratios relative to the standard reference (LNCap) were generated for all the genes for each experiment. Only genes that were expressed at a significant level above the background in all the experiments were used in the analysis. This intensity cutoff across all the samples (4 primary tumors, 7 time points, and 4 recurrent tumors) resulted in 2648 genes that were expressed at a sufficient level to give reliable data.

Differences in gene expression between primary CWR22 and recurrent CWR22R xenografts were measured in four separate experiments. Direct comparisons of a pool of four primary xenografts and four recurrent tumors were done by labeling one pool with Cy 5 and the other with Cy 3 and hybridizing them together (Direct P/R column). The experiment was repeated with the Cy 3 and Cy 5 dyes reversed. Each of the four primary and four recurrent xenografts were also hybridized individually against the standard reference cell line (LNCap) in eight independent experiments on a different microarray print of the same clones. In Table 4, the average of the four recurrent ratios relative to LNCap is divided by the average of the four primary recurrent ratio relative to LNCap, to give the column labeled Avg(4xL)/Avg(4xL). Finally, the pools were also hybridized separately on two different cDNA microarray slides against a standard reference (LNCap) to give the last column in Table 2, labeled Direct 4x4 Pooled. Table 2 shows genes that were selected based on consistency across all the pooled experiments and a significant difference in the average of four primary to four recurrent in the eight independent experiments. (Significance was considered at the 99 % confidence level).

Table 4

Name	Image Clone ID	Direct R/P	Avg(4xL)/ Avg(4xL)	Direct 4x4 Pooled
S100 calcium-binding protein P	135221	16.08	4.84	8.70
inhibitor of DNA binding 3, dominant negative helix-loop-helix protein	756405	14.53	2.16	2.48
mitogen-activated protein kinase-activated protein kinase 2	812251	5.86	2.16	2.91
ESTs	139331	4.66	1.91	2.41
cysteine-rich protein 1 (intestinal)	1323448	4.60	1.68	2.19
zinc finger protein homologous to Zfp103 in mouse	768562	3.77	2.53	2.81
forkhead (Drosophila)-like 13	811600	3.11	1.76	3.67
signal transduction protein (SH3 containing)	795730	3.34	2.38	2.90
thyroid hormone receptor, alpha (avian erythroblastic leukemia viral (v-erb-a) oncogene homolog)	795330	3.16	1.63	1.57
enigma (LIM domain protein)	502682	3.05	1.76	1.89
CDP-diacylglycerol synthase (phosphatidate cytidylyltransferase) 1	135630	2.35	2.32	2.77
Phosphoribosyl pyrophosphate synthetase 2	503097	2.33	2.11	2.52
proliferating cell nuclear antigen	789182	2.50	2.09	1.67
ESTs	108351	2.47	1.96	1.93
Human insulin-like growth factor binding protein 5 (IGFBP5) mRNA	45542	0.36	0.43	0.45
EST	866702	0.34	0.35	0.41
potassium intermediate/small conductance calcium-activated channel, subfamily N, member 4	756708	0.39	0.33	0.47
ESTs	470279	0.33	0.42	0.73
MUF1 protein	738900	0.50	0.32	0.69
thrombospondin 1	810512	0.31	0.54	0.39
dipeptidylpeptidase IV (CD26, adenosine deaminase complexing protein 2)	343987	0.43	0.38	0.29
diacylglycerol kinase, alpha (80kD)	815555	0.29	0.59	0.53
3-hydroxy-3-methylglutaryl-Coenzyme A synthase 2 (mitochondrial)	757222	0.29	0.69	0.35
protease inhibitor 12 (neuroserpin)	564621	0.27	0.69	0.42
EST, collagen, type IV, alpha 5 (Alport syndrome)	42864	0.25	0.28	0.32
crystallin, mu	42373	0.35	0.42	0.20
nuclear VCP-like	416390	0.19	0.61	0.58
Homo sapiens clone 23568, 23621, 23795, 23873 and 23874 mRNA sequences	144747	0.19	0.47	0.50
Human isolate JuSo MUC18 glycoprotein mRNA (3' variant), complete cds, fibronectin 1	897531	0.18	0.17	0.18
surfactant, pulmonary-associated protein A1	841507	0.17	0.41	0.43
solute carrier family 12 (sodium/potassium/chloride transporters), member 2	685801	0.14	0.48	0.23
membrane fatty acid (lipid) desaturase	324891	0.13	0.35	0.45
Homo sapiens eIF-2-associated p67 homolog mRNA, complete cds	39093	0.04	0.48	0.42

A hierarchical, unsupervised approach was first used to cluster the data. Although it was difficult to identify clusters with the small number of samples in the time course, a cluster containing 135 genes that decreased in gene expression after therapy and then were re-expressed after recurrence was identified (listed in Table 5; 143 sequences are listed, eight of which are duplicates). Other genes that decreased and were subsequently re-expressed were also observed, but did not cluster together because of differences in kinetics.

Template clustering (as described above) was developed and used to organize the cDNA microarray data according to expression kinetics during the course of therapy from primary to 16 days following castration. This is a supervised clustering approach, in which a correlation of each gene to a set of templates selected to reflect the temporal nature of the phenotype is calculated. Templates with the best (maximum) expression profile were then utilized to calculate a ranking (cluster location), to sort the genes based on their kinetics. The recurrent (R) time point was not used to calculate correlation to the templates. Color-coding was used to reflect the change that occurs in expression (not ratio) during the therapy, red being the maximum point and green the minimum point.

A database of the gene identification information and template clustering parameters such as cluster location (sorting rank), maximum correlation coefficient, and calculated fold change from lowest to highest point of expression during the first six time points was constructed. This database facilitates mining for HRPC-related genes by variable stringency query-based searches based on (1) amplitude of change during therapy, (2) specifics of kinetics such as early or late increase or decrease, and (3) extents of correlation to the phenotype templates. Filtering for higher "maximum correlation coefficient" allows the (1) selection of profiles that are more strongly associated with the therapy and (2) the elimination of noise in the data generated by large expression ratio differences due to tumor differences or experimental artifact. The genes were sorted and plotted according to the template that their expression most closely resembled. Since the order of each gene is not dependent of the order of other genes, filtering of the data and re-clustering does not require new calculations.

Unsupervised gene clustering was first used to find genes with similar gene expression profiles. The plot shown in FIG 1A illustrates all 2648 genes that showed sufficiently high expression levels to be used in this analysis, organized in hierarchical clusters, as demonstrated by the dendogram on the left. Therapy time points are (from left to right) numbered 1 through 7 and represent primary (P), 0.5 days, 2 days, 4 days, 8 days, 16 days post castration and recurrent (R), respectively; genes are stacked vertically. A group of 139 genes that decrease after castration and then are re-expressed in the recurrent tumors forms a definable cluster (as indicated by the close branching in the dendogram); this group is outlined by a rectangle. Grey-coding/shading in FIG 1A reflects actual ratio to the reference as indicated by the key below the cluster.

Template clustering followed by filtering for greater than two-fold ratio difference during therapy response and for profiles that have at least a 0.7 maximum correlation coefficient to any of the 12 templates resulted in 604 genes (listed in Table 1). The plot in FIG 1B illustrates supervised-template based clustering of these 604 genes. The order of sorting is determined by the template for

which each genes' expression is best correlated to, as indicated on the left of the cluster and described in the methods. Color-coding is usually used to represent the relative transcript expression ratio, as measured by cDNA microarray analysis. Red customarily indicates the maximum point in gene expression, green the minimum, and levels closer to the mean approach black. These colors have been converted to shades of grey, as shown in the key below the cluster. The 604 genes are stacked/clustered vertically for each of the time points in the experiment, organized from left to right and labeled 1 through 7 as for FIG 1A. For each of the first six time points, a correlation coefficient to each of the 12 templates was calculated for the expression profiles of the 2648 sufficiently expressed genes. The average of the three maximum correlation coefficients was used to calculate a precise cluster location that reflects the association of that gene to a particular profile, represented by a continuum of templates guided by the 12 shown in FIB 1B.

Temporal Gene Expression Program Associated with the Response to, and Failure of, Hormone Therapy

Filtering data from the microarray analyses at the level of a three-fold difference in the regressing time points yielded a set of 131 genes (Table 2), 59 of which clustered together due to their correlation (>0.8) to decreasing templates (labeled "Decreasing" in Table 2; Image ID Clone numbers: 788256, 196303, 415089, 785707, 774446, 781047, 126650, 795936, 131316, 700792, 324891, 811015, 207288, 204257, 769921, 249603, 207358, 435076, 43550, 416833, 814701, 898286, 204214, 796646, 129865, 66406, 451907, 711768, 416833, 453107, 509887, 66728, 789204, 626716, 47833, 149013, 531319, 789182, 856427, 725454, 49352, 293727, 273546, 53316, 42059, 855487, 281003, 898062, 24145, 134719, 684655, 29063, 45233, 814117, 283315, 785778, 840567, 767817, 742132) and FIG 1C, and 72 of which clustered together due to their correlation to increasing templates (labeled "Increasing" in Table 2; Image ID Clone numbers: 843249, 298417, 815284, 839101, 153411, 243816, 489839, 39920, 343867, 503083, 897667, 66322, 195340, 78294, 630013, 257162, 813841, 266146, 840511, 415084, 841695, 137139, 136557, 509731, 840460, 111750, 711918, 809998, 784296, 82734, 322160, 177772, 223350, 502832, 813611, 140806, 772425, 246430, 132140, 137096, 768246, 897906, 1412481, 124071, 42070, 362483, 382195, 130826, 811162, 796613, 138936, 811088, 142788, 345935, 773301, 781139, 810960, 813823, 242062, 843098, 51582, 839991, 840687, 66731, 272327, 121792, 120964, 1374571, 842836, 360885, 815774, 35828). On the right of each gene expression profile color plot is a number that corresponds to the fold change (Δ) in ratio between the first six time points. The gene cluster order was determined by the order of templates and cluster location, as described. Gene identifiers shown in FIG 1C include IMAGE clone ID and the current unigene cluster number, name and description. Genes that have previously been reported to be direct targets of the androgen receptor are shown in bold text, and include the following: malate dehydrogenase 1, NAD (soluble) (MDH1), proliferating cell nuclear antigen (PCNA), brain-specific alpha tubulin (TUBA3), ornithine decarboxylase 1

(ODC1), lactate dehydrogenase A (LDHA), a disintegrin and metalloproteinase domain (ADAM9), v-fos FBJ murine osteosarcoma viral (FOS), and andromedulin (ADM).

The template based, supervised cluster of 59 genes (filtered for greater than three-fold change; greater than 0.8 maximum correlation coefficient; only decreasing templates) (listed in Table 2, and labeled "Decreasing"), representing the genes with the largest decrease after castration, had extensive overlap (51 of 59 genes in common, Image Clone ID numbers 767817, 840567, 785778, 283315, 814117, 45233, 29063, 684655, 134719, 24145, 898062, 855487, 42059, 53316, 273546, 293727, 49352, 725454, 856427, 789182, 531319, 149013, 47833, 626716, 789204, 453107, 416833, 711768, 451907, 66406, 129865, 796646, 204214, 898286, 814701, 416833, 43550, 435076, 207358, 769921, 204257, 207288, 811015, 700792, 131316, 795936, 126650, 781047, 774446, 785707, 415089) with the hierarchical (unsupervised) cluster of 139 genes (Table 5). However, the unsupervised cluster was not inclusive of all the genes that responded to the therapy (since it only contained 139 of the 305 genes with a profile that fit a decreasing template with a minimum two-fold difference and >0.7 max. correlation coefficient). Furthermore, although supervised clustering did identify that at least 74 genes increased by more than three-fold and that fit an increasing template with more than 0.8 correlation coefficient, it was difficult to identify a coherent unsupervised cluster of increasing genes.

By template based clustering and filtering the data, a temporal gene expression program (fingerprint), or cluster of genes, was identified that had the largest expression decrease after castration and the best correlation to a decreasing temporal template (FIG 2). The genes are plotted from early repressed genes on the top, and gradually being repressed at later time points down the list to the bottom genes that had a late onset repression. Investigation of the genes in this list of 59 revealed at least eight genes previously known to be stimulated by androgens, and probably direct targets of the AR. The identification of these AR responsive genes in this cluster further substantiates the utility of template based gene clustering in identifying therapy response associated genes and suggests that other genes in this list may be previously unknown AR responsive genes.

Further examination of the genes in this cluster revealed that it is very rich in several important cell cycle regulators. These include genes known to be associated with cell growth of prostate cancer including PCNA, ornithine decarboxylase 1, c-fos, and tubulin. Most of the genes in this cluster however, are novel cell cycle regulators that were not previously associated with androgen ablation in prostate cancer. These include the following (Image ID Clone numbers in parentheses):

two **BUB** (budding uninhibited by benzimidazoles) genes, which regulate the cell cycle at the mitotic checkpoint by controlling chromosome segregation and responding to spindle disruption (781047, 785778, 842968);

UBCH10, a cyclin-selective ubiquitin carrier that regulates the destruction of mitotic cyclins (769921);

CDKN3, a CDK-2 associated dual phosphatase (700792);

CDC2 delta T which regulates entry into S-phase and mitosis (898286);

CDC18L, which initiates replication (204214);

CKS2, a kinase that activates CDC28 (725454);

MAD2L1, which regulates mitotic checkpoints especially sensitive to kinetochore and spindle loss (814701);

CENPF, a centromere/kinetichore cell cycle protein (435076);

STK12, a chromosome associated kinase that plays an important role in centrosome duplication regulation, aneuploidy, and amplification (531319);

NEK2 a protein kinase that regulates G2-M transition (415089);

CDC20, responsible for nuclear movement prior to anaphase and chromosome separation (898062); and

CDC45L, required for the initiation of DNA replication (453107).

The abundance of growth regulatory genes in this cluster, and of genes known to be direct targets of the androgen receptor, provides further supports that genes in this cluster are dependent on androgens and are mediating the AR dependent growth arrest following androgen ablation.

Transcript levels of the genes in this cluster are restored when therapy fails, suggesting that these are also the genes that mediate the androgen-independent growth in recurrent tumors. These observations are also consistent with the hypothesis that resistance to therapy occurs through an androgen independent activation of the AR.

In addition, several genes were repressed that have never previously been associated with cellular proliferation. FKBP5, for example, was repressed by as much as 5.8-fold after castration. This may be a direct effect of decreased androgen receptor transcriptional activation. FKBP5 has been associated with the glucocorticoid receptor, and targeting of FKBP proteins has been shown to lead to deregulation of several signal transduction pathways.

Another gene that showed a large amplitude change after castration, with unknown consequence, is transmembrane 4 superfamily member 1 (7.1 fold decrease). Conversely, transmembrane 4 superfamily member 3 increased after castration (3.2 fold). Putative signaling molecule serine/threonine kinase 12 (7.0 fold decrease) and insulin induced gene 1 (8.1 fold decrease) also showed substantial expression level changes after castration. Like the known cell cycle regulators, the expression of all these other genes is restored in the recurrent tumors. It is likely that these genes mediate growth arrest after therapy, and tumor re-growth after development of therapy resistance, and therefore these genes are ideal drug target candidates.

In addition, some important genes that changed but did not make the top 59 list include S100P, ID3, PSA and c-myc mRNA, which decreased by 5.2, 2.85, 2.77 and 3.01 fold respectfully during regression (FIG 2). These were not included in the primary list either because they did not meet the 3 fold cut-off, or because the maximum correlation coefficients were less than 0.8 (0.51

0.67, 0.71 and 0.50). Of this group, S100P and ID3 are especially good candidate drug targets because they are also over-expressed in recurrent CWR22R relative to their primary counterparts (Table 2).

Most, but not all, of the genes that show increased expression following therapy response generally remained elevated in the recurrent tumors. This is contrary to the repressed genes, whose transcript levels were largely restored. Some genes, however, increased during therapy and then were restored in the recurrents. This group includes: the UDP glycosyltransferase 2 family, polypeptide B15 and UGT2B4, sialyltransferase 1 (beta-galactoside alpha-2,6-sialytransferase), fatty-acid-Coenzyme A ligase, human metallothionein (MT)I-F gene, tumor suppressor PTEN, cadherin 3, placental-cadherin, gelsolin (amyloidosis, Finnish type), TAP binding protein (tapasin), and several other transcripts. The increase in PTEN indicates that the AKT S6 kinase pathway may be inhibited following castration, suggesting that therapeutic intervention with rapamycin may mimic this inhibition in recurrent tumors.

EXAMPLE 2

Identification of Further Genes with Altered Expression in Hormone Refractory Prostate Cancer

Using different microarrays, and methods essentially similar to those described above in Example 1, additional HRPC-related nucleic acid molecules were identified and further characterized. These HRPC-related nucleic acid molecules also show differences in expression during prostate cancer development, and particularly during hormone ablation therapy and subsequent progression to a hormone-refractory condition.

Methods and Material:

Methods and materials were essentially as described in Example 1, except that additional custom cDNA microarrays were used, constituting 6605 to 8000 elements (sequence verified clones from Research Genetics, Huntsville, Alabama), representing different (non-redundant) transcripts including 4032 to 7700 known (named) genes (Mousses *et al.*, *Funtional Genomics: Gene Expression Analysis by cDNA Microarrays* Livesey and Hunt (eds). Oxford University Press: Oxford, pp. 113-137, 2000.). All xenografts were analyzed at least twice. Either LNCap or CWR22R were used as a reference and labeled with Cy5. The reference cDNA was simultaneously hybridized with Cy3 labeled test specimens on a cDNA microarray as previously described (Mousses *et al.*, *Funtional Genomics: Gene Expression Analysis by cDNA Microarrays* Livesey FJ and Hunt SP (eds). Oxford University Press: Oxford, pp. 113-137, 2000.). Fabrication of the microarray slides, image generation, and the software used for the ratio analysis, and bioinformatics were as described above. Mousses *et al.*, *Funtional Genomics: Gene Expression Analysis by cDNA Microarrays* Livesey and Hunt (eds). Oxford University Press: Oxford, pp. 113-137, 2000.

Template-based clustering was performed as described above.

Most Systematically Altered Genes

Another set of genes was identified that showed differential expression between primary and recurrent tumors. Based on the mean gene expression ratios from six recurrent and four primary tumors, expression levels of 104 of the 3495 informative genes (3.0%) were significantly (2-fold or more) increased, and those of 60 genes (1.7%) decreased in the recurrent tumors. FIG 3A shows 30 genes (out of a total of 164 differentially expressed genes) that were most systematically altered in the recurrent tumors. These genes include SCYD1, S100P, CCND1, CRIP1, ISG15, SCNN1A, ZFP103, MAPKAPK2, UTG2B15, RABGGTA, NFKBIA, SLCYA5, AP3B2, PTPN2, FOXJ1, and APOC1 (all upregulated) and FLJ23538, OXCT, PFKP, TNRC3, HXB, PFKP, OAT, PFKP, RFP, THBS1, LMO4, MLD, CRYM, MME, HMGCS2, and SLC12A2 (all downregulated).

Among the 164 genes were several genes coding for proteins that either converged on the PI3K/AKT/FRAP pathway or represented direct targets of macrolide drugs (such as rapamycin and FK506). As highlighted in FIG 3B, several genes that were androgen-responsive and re-expressed in the recurrent tumors (CCND1, ODC1, EIF1EBP1, MAPKAPK2, NFKBIA, CDS1, FKBP4, and FOXJ1) met these criteria and suggested involvement of rapamycin-sensitive signaling in hormone-refractory tumors.

These findings appeared to indicate that rapamycin-sensitive gene products and signaling pathways play a role in androgen independent growth in the recurrent tumors. To further evaluate this hypothesis, the effects of rapamycin and FK506 on the growth and viability of a cell line established from the recurrent CWR22R xenografts were studied. Rapamycin is a known inhibitor of the PI_3K/AKT/FRAP pathway (Kunz *et al.*, *Cell*, 73:585-596, 1993; Brunn *et al.*, *EMBO J.*, 15:5256-5267, 1996; Sekulic *et al.*, *Cancer Res.*, 60:3504-3513, 2000), and FK506 targets many of the same intracellular proteins as rapamycin. Death of the hormone-independent CWR22R cells was observed at very low doses of rapamycin (IC50 ~ 0.1 nM) (FIG 3D), whereas hormone-responsive LNCap prostate cancer cell lines exhibited partial inhibition, even at high doses (FIG 3D). FK506 treatment did not have an inhibitory effect on either the CWR22R or LNCap cells even at the highest doses tested (greater than 80% cell survival at a dose of 10 mM). The results are based on two different cell lines that are not isogenic and may have other differences contributing to the observed effects. However, both these results of the global-scale gene expression studies and the data from the in vitro sensitivity testing, indicate that further studies are warranted to explore rapamycin as a candidate drug for the treatment of hormone refractory prostate cancers.

Cancer cells exhibit greater than a 1000-fold (IC_{50} ranging from <1 nM to >10 mM) variability in their sensitivity to rapamycin, possibly reflecting mechanisms of intrinsic resistance (Hosoi *et al.*, *Mol. Pharmacol.*, 54:815-824, 1998). Cancer cells that have activated genes and pathways that signal through the PI3K/AKT/FRAP pathway may be particularly sensitive. For example, IGF-1 receptor activation is associated with the efficacy of rapamycin treatment in childhood sarcomas (Dilling *et al.*, *Cancer Res.* 54:903-907, 1994). Several growth factors and related genes that we observed to be overexpressed in the recurrent prostate cancers relative to the

primary tumors (such as HGF, VEGFC, FGF2, IGFBP3, PDGFA, LTBP4, GFR, PGF, ITPKB, CDS1, and FKHL13) could have similarly contributed to the activation of the PI3K/AKT/FRAP pathway and alterations in the rapamycin target expression.

Finally, the two macrolide drugs rapamycin and FK506 bind similar intracellular targets but have different biological effects in hormone-refractory prostate cancer. These differences may be informative in elucidating those molecular pathways that are most critical for progression of prostate cancer. Rapamycin and FK506 both bind to FKBP12 (FK506-binding protein 12) (Sabers *et al.*, *J. Biol. Chem.*, 270:815-822, 1995; Liu *et al.*, *Cell* 66:807-815, 1991). Rapamycin-FKBP12, but not the FK-506-FKBP12 complex, inhibits FRAP (FKBP-Rapamycin Associated Protein), a member of the phosphoinositide-3-kinase related kinases that regulate translation following mitogenic activation of the PI3K/AKT/FRAP pathway. In contrast, FK506, but not rapamycin, inhibits calcineurin activity (Liu *et al.*, *Cell* 66:807-815, 1991). This suggests that, of the many known and unknown targets of rapamycin and FK506, FRAP and the activity of the PI3K/AKT pathway is a more likely candidate than calcineurin as a drug target in hormone-refractory prostate cancer.

This example clearly illustrates that transcriptional profiling can be used to identify candidate drugs for treatment of prostate cancer, and this approach generally, as well as the present findings more specifically, can be used for a basis of such treatment decisions.

EXAMPLE 3

Analysis of Specific Genes

A direct comparison of a pool of four primary CWR22 xenografts and four recurrent CWR22R xenografts was done by labeling one pool with Cy5 and the other with Cy3 and hybridizing them together (Direct P/R column in Table 2). This resulted in 251 genes (3.8% of the 6605 genes assayed; listed in Table 6) that were differentially expressed at the 99 % confidence level. This analysis was also done against the standard reference for each tumor individually and in pools with the most consistently differentially expressed genes shown in Table 4.

One of the most highly differentially expressed genes is a calcium binding protein, S100P. It was found to be expressed 16 times (by cDNA microarray analysis) to 100 times (by Northern hybridization analysis) higher in one recurrent xenograft compared to the primary. The S100P protein has been reported to be associated with increased survival and loss of senescence in breast cancer cells. This data indicates that S100P expression may be androgen dependent, as would be expected if it is involved in prostate cancer progression.

Several immunophilin-like proteins were also identified as being differentially expressed. FKBP5, in addition to being overexpressed by about two-fold on average, is one of the most repressed genes after castration. During recurrence, its expression is restored to higher levels than in the primary. FKBP5 is a member of the large immunophilin chaperone proteins, which have been shown to interact with HSP90 and several steroid receptors. The expression of this protein not only appears to be regulated by the androgen receptor function, but also may affect androgen receptor

activity by protein folding of the nascent receptor or by modulating its binding affinity to ligands. There are several inhibitors (*e.g.*, FK506 and rapamycin) that bind to immunophilins, resulting in either calcineurin inactivation and or the inhibition the phosphotidylinositol 3-kinase/PTEN/AKT/FRAP pathway (Zhong *et al.*, *Cancer Res.* 60:1541-1545, 2000).

The phosphorylated substrates of this pathway include calcineurin and ties into the calcium signaling pathway. In addition, IkB (which regulates NFkB), NFAT, and BAD are each substrates for this pathway and are all involved in regulation of cell survival. Decreased expression of PTEN, and increased expression of CDP-DG synthase, IkB, PHYH, and several other changes also converge on, and possibly alter activity of, this pathway. Drugs that target immunophilins such as FK506 and rapamycin have been shown to inhibit this pathway at the level of FRAP, leading to (1) loss of activity for kinases with mitogen activated protein kinase (MAPK) like substrates and (2) inactivation of calcineurin. Differential gene expression data disclosed herein indicate that such drugs such as rapamycin and FK506 could have a dual role in preventing androgen independent progression of prostate cancer, by both (1) blocking signal transduction from the phosphotidylinositol 3-kinase/PTEN/AKT/FRAP pathway and (2) interfering with androgen receptor protein folding and assembly. This is an example of the differential gene expression discussed herein, to assist in selecting new therapies for treatment of primary and recurrent (hormone-refractory) prostate cancer.

EXAMPLE 4

Tissue Microarray Analysis of Candidate Biomarkers

This example provides in-depth analysis of several HRPC-related genes, including illustrations of the clinical relevance of these genes in prostate cancer progression and staging. High throughput molecular validation of candidate genes in clinical specimens was accomplished by using tissue microarray technology to assess the utility of these HRPC-related genes as biomarkers and drug targets. Using a tissue microarray in this fashion represents an important method to cross-validate data from experimental systems and human cancer specimens.

Tissue microarray methods were carried out essentially as described above, and as known in the art; see, for instance, Kononen *et al.*, *Nat Med.* 4(7):844-847, 1998) Clinical translation of novel gene products where an antibody does not exist can be detected on tissue microarrays using isotopic in situ hybridization (ISH) (Kononen *et al. Nat Med.* 4(7):844-847, 1998; Frantz *et al.*, *J Pathol.* 195(1):87-96, 2001)

S100P

The prevalence of S100P protein overexpression was investigated by immunohistochemistry, in 440 human prostate cancer specimens at various stages of progression. These specimens were arrayed in a prostate cancer progression tissue microarray (Bubendorf *et al.*, *J Natl Cancer Inst* 91:1758-1764, 1999 and Bubendorf *et al.*, *Cancer Res.* 59:803-806, 1999). This

array also contained about 50 different prostate cancer xenograft samples, including those used in the cDNA microarray experiments.

S100P mRNA was measured by three different methods in nine xenografts. cDNA microarray ratios measure the expression of S100P transcript by the amount of cDNA hybridized relative to the standard reference. Northern analysis with a PCR amplified fragment of the S100P against a blot of the same RNA used in the cDNA microarray analysis produced a fragment of expected size (~0.5 kb). Northern hybridization bands were quantified using ImageQuant software from a scanned autoradiogram. An mRNA *in situ* hybridization (ISH) was performed by radio-labeling eight non-overlapping oligonucleotide (~45 bp) that span the coding region and hybridizing them to tissue microarrays containing hundreds of sections (including xenografts) described in the methods and materials. The signal was quantified using a Fuji phosphoimager and scanner and Bos software. The quantification of each of these there methods is plotted above the images for each of the nine xenografts. The absolute values are normalized to three of the primary tumors with the lowest Northern hybridization levels. For each of the xenograft tumors, the S100P protein expression is shown by IHC staining is shown below the graph (FIG 4).

FIG 4 shows that in at least the xenograft samples there is good concordance between Northern hybridization, cDNA microarray, and mRNA *in situ* on tissue microarray quantitation of S100P transcript levels. These mostly but not always correlate with immunohistochemical staining. In at least a few cases, higher protein expression was observed with moderate levels of mRNA, indicating possible post-transcriptional regulation.

In situ mRNA hybridization was also used to quantitatively measure transcript levels on tissue microarray sections. Immunohistochemical analysis of S100P protein expression in 440 human prostate cancer specimens at various stages of progression is shown in FIG 5. An S100P-specific antibody was used to stain prostate tissue sections on a tissue microarray. The staining intensity was scored by two pathologists, using a scale of from 0 to 4. The results in FIG 5 show the percentage of cancers at each stage of prostate cancer progression that had strong staining (score of 3 or 4). FIG 5 shows that the high expression of S100P protein is associated with progression in clinical prostate cancers, with increasing expression in refractory and metastatic disease.

FKBP5

Translation of the observations on FKBP5 to clinical specimens is of interest because of this protein is associated with therapeutic response, and is over-expressed in recurrent tumors. Until now, it was thought that FKBP5 was only expressed in T-cells, and that it would make a good drug target for specific immunosuppression through the inhibition of glucocorticoid receptor transcriptional activation. Using prostate cancer progression tissue microarrays, FKBP5 was found to be expressed specifically in secretory cells of the normal prostate and in prostate cancer cells, but not in supporting stromal cells. Analysis of FKBP5 protein expression by IHC on the same prostate cancer tissue microarray as discussed above indicated that FKBP5 is expressed in the majority of prostate cancers,

but an association with progression was not observed. Many of the primary and early lesions had common expression of this protein, thereby indicating that FKBP5 would not make a good biomarker for prostate cancer progression or the development of hormone refractory or metastatic disease. However, FKBP5 down-regulation does appear to be associated with therapeutic response, making it a candidate for therapeutic targeting in a large percentage of clinical tumors.

LMO4 and *CRYM*

LMO4 and *CRYM* genes were substantially down-regulated in the CWR22R tumors relative to primary CWR22, for mRNA ISH studies. In both cases, mRNA ISH on TMAs validated the relative expression levels seen by cDNA microarrays in the CWR22 xenograft specimens. This analysis revealed a lower level of *LMO4* and *CRYM* expression in 17 recurrent CWR22R xenografts ($p<0.001$) as compared to 19 primary CWR22 xenografts. In addition to permitting us to validate our observations, the xenografts on the tissue microarrays were also used to compare the measurement of mRNA by cDNA microarray and mRNA ISH on a tissue microarray. As an example, there is a high correlation ($r=0.96$, $n=16$) between the levels of LMO4 mRNA measured by mRNA ISH on a tissue microarray and data from cDNA microarrays.

A significant decrease ($p<0.001$) of mRNA levels was observed for both *LMO4* and *CRYM* during tumor progression in cancer patients by mRNA ISH on the TMA. The mean intensity of actin mRNA was used as a negative control in the mRNA ISH. Comparison of mRNA ISH levels between primary and hormone refractory tumors on the same array revealed no significant differences between the two groups ($P = 0.927$).

Since antibodies are often not available for gene products discovered from cDNA microarray surveys, it remains essential to detect these transcripts on tissue microarrays using mRNA ISH. We validated here mRNA ISH-based detection of transcripts by inserting into the TMAs specimens that were originally used in the cDNA microarray analyses. There was an excellent correlation between mRNA ISH and cDNA microarray results, indicating that this method can be used to accurately measure mRNA levels in samples on a tissue microarray format. mRNA ISH was performed with several radioactively labeled oligonucleotide probes for different regions of the target genes. The use of short probes to different regions of the genes made it possible to obtain a signal even from degraded mRNAs that inevitably exist in clinical specimens. CRYM and LMO4 were down-regulated in clinical specimens from hormone-refractory tumors, which is in line with the cDNA microarray results in the CWR22 xenograft model system.

LMO4 is a member of the LIM-only (LMO) subfamily of LIM domain-containing transcription factors that is expressed during embryonic development (Kenny *et al.*, *Proc. Natl. Acad. Sci.* 95:11257-11262, 1998) and Crystallin mu (*CRYM*) codes for a thyroid hormone binding protein (Kim *et al.*, *Proc. Natl. Acad. Sci.* 89:9292-9296, 1992; Aoki *et al.*, *J. Invest. Dermatol.* 115:402-405, 2000). Both had transcript levels that were negatively associated with clinical progression. A role in prostate cancer progression has previously not been reported for either of these genes. It is believed

that the observations presented herein indicate that perturbation of these genes has a functional role in clinical prostate cancer progression and pathogenesis.

This example illustrates tissue microarray technology validation of the *in vivo* involvement of four new prostate cancer related genes. Alterations in S100P, FKBP5, CRYM and LMO4 genes are not only involved in the acquisition of androgen-independent growth and failure of therapy in prostate cancer xenografts but also with the progression of cancer in patients.

EXAMPLE 5

Targeting Candidate Genes with Known Drugs

This example demonstrates the clinical effectiveness of selecting drug targets and genetic markers, indeed entire metabolic pathways, using the herein-disclosed HRPC-related genes. Several drugs were identified based on their known interaction with one or more of the HRPC-related genes or implicated pathways, and the activities of these drugs in controlling prostate cancer cell growth was examined.

Cell viability and Drug Treatment

Exponentially growing LNCaP or CWR22R cells were trypsinized and plated at 0.5×10^5 cell/ml or 1×10^5 cell/ml respectively in 96-well culture plates. After 24 hours, cells were treated for 72 hours with serial twofold dilutions of compound. DMSO was added to the control wells. Cell viability was measured by the WST-8 assay (Dojindo Molecular Technologies Inc.). The WST-8 [2-(2-methoxy-4-nitrophenyl)-3-(4-nitrophenyl)-5-(2,4-disulfonyl)-2H tetrazolium, monosodium salt] assay is based on the conversion of the tetrazolium salt WST-8 to highly water soluble formazan by viable cells (Tominaga *et al.*, *Anal. Commun.* 36, 47-50, 1999). The WST-8 reagent solution was added to each well. After incubation for three hours at 37 °C, the absorbance was measured at 450 nm with a reference wavelength at 630 nm. The experiments were performed in triplicate. The data are representative of three separate experiment.

MS-275 and TSA

A literature search, coupled with a search of previous drug treatment data, was used to identify known compounds that could be used to target one or more of the 604 genes that changed at least two-fold following therapy response (FIG 1 and Table 1), or one or more of the 251 genes that were differentially expressed between primary and recurrent tumors (Table 6).

ID3 (clone: 756405) has recently been shown to be required for angiogenesis (Lyden *et al.*, *Nature* 401:670-677, 1999). The inventors have also observed a decreased in thrombospondin (clone: 810512) (an angiogenesis inhibitor) during prostate cancer progression, suggesting that the expression of these two genes is changed in opposite directions in recurrence to achieve the same biological outcome, increased angiogenesis. Currently, there are no known inhibitors of ID3 (clone:

756405), but the inventors have observed in an independent set of cDNA microarray experiments that TSA, induced thrombospondin (clone: 810512) by as much as 8.6 fold in PC3M cells *in vitro*.

TSA works by an unknown mechanism, possibly by histone deacetylase inhibition resulting in altering transcription of a large number of genes. TSA treated PC3M cells revealed targets that were similarly affected in the direction of the growth arrested xenografts. Both drugs reduced kallikrein 3 (prostate specific antigen) (clone 824568) by two-fold, possibly reflecting an inhibition of AR-dependent transcriptional activation. Histone acetyltransferase 1 (clone: 745360) and acetyl-Coenzyme A acyltransferase (clone: 27848) are both decreased by about two-fold after castration, indicating that histone deacetylase inhibition might mimic this effect (growth suppression).

Cyclin D1 (clone 841641) mRNA levels dropped to about 50 % only slightly after castration but the CWR22R recurrent tumors overexpressed it relative to the primary. Similarly chromosome condensation 1 (clone 724615) was 2.8 times higher (pooled experiment) in recurrent tumors. Both Cyclin D1(clone 841641) and chromosome condensation 1 (clone 724615) were repressed by about three-fold by TSA treatment. The recurrent to primary ratio for protease inhibitor 12 (neuroserpin) (clone 564621) was 0.27, but TSA induced it by 8.67 fold.

Gene expression changes in response to treatment with these two drugs indicated that they might restore the expression of several genes that are associated with therapy resistance in CWR22R xenografts. TSA effectively inhibited growth of CWR22R, as indicated in FIG 3D. It is not known which of the above mentioned targets were affected, or by which mechanisms these two drugs caused growth arrest. It is possible that these drugs had a more global gene expression effect, which simultaneously restored multiple androgen responsive genes that are required for growth in the recurrent tumors.

Rapamycin and FK506

Sirolimus (Rapamycin) and Tacrolimus (FK506) are bacterial macrolides that are produced by fungi to suppress the growth of competing organisms. These drugs are immunosuppressants used extensively to prevent organ rejection. Although the two drugs are very similar both in structure and in their cellular targets, known as immunophilins (also called FKBP for FK506 binding proteins), the mechanism by which they cause immunosuppression is different. FK506 binds to immunophilins and the complex inhibits calcineurin in T-cells. In contrast, rapamycin-immunophilin complex inhibits signaling of the S6-kinase (clone: 204148, which also responds to castration) causing cell cycle arrest in T-cells. In addition, there are "macro" immunophilins that have been found to interact with steroid receptors, which may work though yet another mechanism to inhibit growth when complexed with these drugs.

Several drug targets identified in this study are involved in immunophilin pathways, suggesting that either FK506 or rapamycin may cause a growth inhibition of hormone refractory prostate cancer. The first such candidate is a macroimmunophilin called FKBP5 (clone: 416833), one of the most strongly repressed genes in primary prostate CWR22 tumors after castration (FIG 2). The

expression of FKBP5 (clone: 416833) is restored in hormone refractory CWR22R prostate cancer. In some tumors, FKBP5 mRNA expression (determined using cDNA microarray and RT-PCR quantitation) is restored to levels higher than found in the primary tumors. The availability of FKBP5 as a drug target was also confirmed using tissue microarray analysis. It is not clear if FKBP5 is required for the proliferation of CWR22R cells, but the expression of the FKBP5 transcript is associated with the proliferation phenotype. FKBP5 is a large protein that associates with steroid receptors, such as the glucocorticoid receptor, through binding to HSP90. It is also possible that FKBP5 interacts with the AR.

Cyclin D mRNA was 2.5-fold higher in a pool of four recurrent tumors compared to a pool of four recurrent tumors. Rapamycin has been shown to target and down-regulate cyclin D protein at both a transcriptional and post-transcriptional level (Hashemolhosseini *et al.*, *J. Biol. Chem.* 273:14424-14429, 1998). Also, p27 had increased after castration by about 2-fold by day 8, and then went back down in the recurrent tumor. Rapamycin can increase p27 levels, making it a candidate for reversing the decrease seen in the recurrent CWR22R. These rapamycin effects on both cyclin D and p27 may be direct, but also may be mediated by inhibition of the phosphotidylinositol 3-kinase/PTEN/AKT/FRAP pathway. Several gene expression changes have been identified herein that could converge to activate this pathway in recurrent tumors, further suggesting that this is a pathway necessary for androgen independent growth. For example, an increased was observed in expression of CDP-diacylglycerol synthase 1 (levels up to 2.77-fold higher in recurrent tumors). CDP-diacylglycerol synthase 1 is a rate limiting enzyme in phosphotidylinositol 3 (PI3) production that has been shown to increase the amplitude and duration of PI3 signaling when overexpressed in model systems. PTEN, which is an inhibitor of this pathway, is increased during regression and re-expressed in the recurrent tumors further illustrating the importance of this pathway for proliferation of recurrent tumors.

It has also been shown that rapamycin inhibits the translation of ornithine decarboxylase (ODC) transcripts by about 50% in epithelial cells. In this study, ODC was repressed (3.8 fold) during CWR22 regression, but then re-expressed in the recurrent CWR22R (FIG 2). Interestingly, FK506 has no effect on ODC transcript levels. Both ODC and cyclin D are important stimulators of proliferation, indicating that rapamycin can be used to target these molecules and cause growth arrest in androgen independent CWR22R cells.

Rapamycin effectively arrested the CWR22R cells *in vitro,* however a complete inhibition was not accomplished at the highest concentration of FK506 (10μM) (FIG 3D). It is believed that the interaction of rapamycin with FKBP5 and its other cellular receptor immunophilins blocks a pathway necessary for growth, while the interaction of FK506 and FKBP5 does not. It is difficult to predict the mechanism by which these drugs exert an effect on a cell, because they bind multiple cellular targets. In this case, several putative cellular targets are known for these two macrolide drugs and at least one, FKBP5, was both associated with the HRPC phenotype, and available in the relevant cells

(FKBP5 protein is expressed in most clinical recurrent tumors). More specific inhibitors of FKBP5 activity can be used to elucidate the role FKBP5 plays in the growth of hormone refractory tumors.

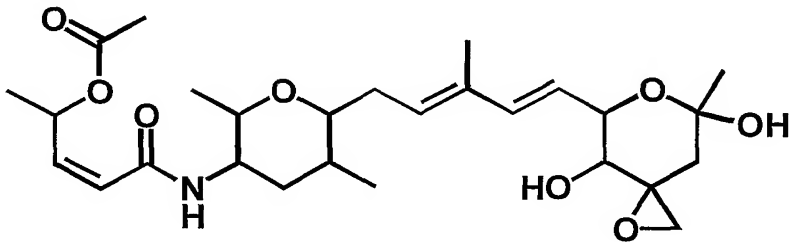
EXAMPLE 6

Pharmacogenomics Analysis

This example illustrates the involvement of gene targets in pharmacological response to various emerging therapies.

Xenografts and Cell lines: Fresh frozen tissue from CWR22 human prostate cancer xenografts (Pretlow *et al.*, *J. Natl. Cancer Inst.* 85:394-398, 199)) was obtained from thirteen different mice at different stages of hormonal therapy and tumor progression (four primary untreated CWR22, five CWR22 therapy time points after 0.5 days, 2 days, 4 days, 8 days and 16 days after castration, and four independent hormone-refractory CWR22R strains). LNCaP (ATCC) and CWR22R (kindly provided by Dr. Jim Jacobberger's Laboratory at Case Western University) cell lines were cultured in RPMI1640 10% fetal bovine serum (Life Technologies Rockville, MD) at 37 °C and 5% CO_2. mRNA was extracted with the FastTrack 2.0 Kit (Invitrogen Corporation; Carlsbad, California).

Drug Treatment and Cell viability: Exponentially growing LNCaP or CWR22R cells were trypsinized and plated at 0.5 x 10^5 cells/ml or 1 x 10^5 cells/ml respectively in 96-well culture plates. After 24 hours, cells were treated for 72 hours with serial two-fold dilutions of either FK-506 (Tacrolimus, Calbiochem Inc., San Diego, California), Rapamycin (Sirolimus) (Sigma Chemical co. St. Louis, Missouri), FR901464 (Fujisawa Pharmaceutical Co., Ltd., Ibaraki, Japan), Trichostatin A - TSA (a histone deacetylase inhibitor; Sigma Chemical co. St. Louis, Missouri) or DMSO as a control. The structure of FR901464 is as follows:



Cell viability was measured (triplicate experiments) by the WST-8 assay (Dojindo Molecular Technologies Inc., Gaithersburg, Maryland). CWR22R cells were treated with various drugs at effective doses for 1, 3, 9 and 24 hours followed by mRNA isolated for cDNA microarray experiments.

Analysis of mRNA expression by cDNA Microarrays: Custom cDNA microarrays were constructed consisting of 6605 to 8000 elements (sequence verified clones from Research Genetics, Huntsville, Alabama), representing different (non-redundant) transcripts including 4032 to 7700 known (named) genes (Mousses *et al.* in *Functional Genomics*, (eds. Livesey & Hunt) 113-137,

Oxford University Press, Oxford, 2000). All xenografts were analyzed at least twice using either LNCap or CWR22R Cy 5 labeled reference cDNA simultaneously hybridized with Cy 3 labeled CWR22 xenograft or CWR22R cell line cDNA on a cDNA microarray according to a previously described protocol (Mousses *et al.* in *Functional Genomics*, (eds. Livesey & Hunt) 113-137, Oxford University Press, Oxford, 2000). Fabrication of the microarray slides, image generation, and the software used for the ratio analysis, and bioinformatics was carried out essentially as described above.

Results:

Trichostatin A (TSA) and FR901464 (an experimental drug found to inhibit the growth of a human solid tumor grown in mice and murine solid tumors; Nakajima *et al.*, *J. Antibiot.* 49:1204-1211, 1996) were selected for *in vitro* testing in CWR22R cells based on previous pharmacogenomics analysis on PC3M cells, which suggested targeting of androgen independent growth associated genes. Rapamycin and FK506 were selected as drugs that also might target some of these candidates. To prioritize candidate gene targets that were not only associated with androgen independent growth but also involved in eliciting an effective drug response, cDNA microarray analysis of CWR22R gene expression was conducted during a time course of drug treatment *in vitro*.

Functional analysis of the dose response of each of these four drugs was carried out using a viability assay on CWR22R cells *in vitro* (FIG 6). A strong inhibition of growth and survival was seen for CWR22R cells with rapamycin, TSA, and FR901464, but not with FK506. Neither FK506 nor rapamycin were as effective at inhibiting the hormone-dependent LNCap prostate cancer cells. The global gene expression profiles indicate mechanisms of drug action that are distinctly different from androgen withdrawal response related signaling (FIG 6A and 6B). Despite this, some candidate genes that were associated with androgen independent growth of CWR22R were also involved in eliciting a response to some of these drug treatments including FKBP5, CRYM, and several others (FIG 6C and 6D; ATP1B2, OAT, QSCN6, GSN, PLU-1, GFPT2, ZCYTOR7, and VDUP1).

Pharmacogenomic analysis revealed that the two drugs TSA and FR901464 work by distinct mechanisms, which do appear not to involve androgen signaling (SM unpublished data). Although some commonly repressed genes were identified across experiments, VDUP1 (upregulated by 1,25-dihydroxyvitamin D-3; accession number XM_002093) was the only transcript that was up-regulated in response to therapy *in vivo* (maximum of 18 fold) and *in vitro* to Rapamycin, TSA and FR901464 (maximum of 73 fold). Based on these results, this gene acts as suppressor of tumor growth and survival.

EXAMPLE 7

Expression of HRPC-related Polypeptides

The disclosed HRPC-related proteins (and fragments thereof) can be expressed by standard laboratory technique. After expression, the purified HRPC-related protein or polypeptide may be

used for functional analyses, antibody production, diagnostics, prognostics, and patient therapy, *e.g.*, for prevention or treatment of prostate cancer (including hormone-refractory or metastatic prostate cancer). Furthermore, the DNA sequences encoding the disclosed HRPC-related proteins can be manipulated in studies to understand the expression of these genes and the function of their products, in particular how these HRPC-related proteins function in the control of or response to hormone-refractory prostate cancer. Mutant forms of human HRPC-related proteins (and corresponding encoding sequences) may be isolated based upon information contained herein, and may be studied in order to detect alteration in expression patterns in terms of relative quantities, tissue specificity and functional properties of the encoded mutant HRPC-related protein. Partial or full-length cDNA sequences, which encode the subject protein, may be ligated into bacterial expression vectors. Methods for expressing large amounts of protein from a cloned gene introduced into *Escherichia coli* (*E. coli*) or other prokaryotes may be utilized for the purification, localization, and functional analysis of proteins. For example, fusion proteins consisting of amino terminal peptides encoded by a portion of the *E. coli lacZ* or *trpE* gene linked to an HRPC-related protein may be used to prepare polyclonal and monoclonal antibodies against these proteins. Thereafter, these antibodies may be used to purify proteins by immunoaffinity chromatography, in diagnostic assays to quantitate the levels of protein and to localize proteins in tissues and individual cells by immunofluorescence.

Intact native protein may also be produced in *E. coli* in large amounts for functional studies. Methods and plasmid vectors for producing fusion proteins and intact native proteins in bacteria are described in Sambrook *et al.* (Sambrook *et al.*, In *Molecular Cloning: A Laboratory Manual*, Ch. 17, CSHL, New York, 1989). Such fusion proteins may be made in large amounts, are easy to purify, and can be used to elicit antibody response. Native proteins can be produced in bacteria by placing a strong, regulated promoter and an efficient ribosome-binding site upstream of the cloned gene. If low levels of protein are produced, additional steps may be taken to increase protein production; if high levels of protein are produced, purification is relatively easy. Suitable methods are presented in Sambrook *et al.* (In *Molecular Cloning: A Laboratory Manual*, CSHL, New York, 1989) and are well known in the art. Often, proteins expressed at high levels are found in insoluble inclusion bodies. Methods for extracting proteins from these aggregates are described by Sambrook *et al.* (In *Molecular Cloning: A Laboratory Manual*, Ch. 17, CSHL, New York, 1989). Vector systems suitable for the expression of *lacZ* fusion genes include the pUR series of vectors (Ruther and Muller-Hill, *EMBO J.* 2:1791, 1983), pEX1-3 (Stanley and Luzio, *EMBO J.* 3:1429, 1984) and pMR100 (Gray *et al.*, *Proc. Natl. Acad. Sci. USA* 79:6598, 1982). Vectors suitable for the production of intact native proteins include pKC30 (Shimatake and Rosenberg, *Nature* 292:128, 1981), pKK177-3 (Amann and Brosius, *Gene* 40:183, 1985) and pET-3 (Studiar and Moffatt, *J. Mol. Biol.* 189:113, 1986). Fusion proteins, for instance fusions that incorporate a portion of a HRPC-related protein, may be isolated from protein gels, lyophilized, ground into a powder and used as an antigen. The DNA sequence can also be transferred from its existing context to other cloning vehicles, such as other plasmids, bacteriophages, cosmids, animal viruses and yeast artificial chromosomes (YACs)

(Burke *et al., Science* 236:806-812, 1987). These vectors may then be introduced into a variety of hosts including somatic cells, and simple or complex organisms, such as bacteria, fungi (Timberlake and Marshall, *Science* 244:1313-1317, 1989), invertebrates, plants (Gasser and Fraley, *Science* 244:1293, 1989), and animals (Pursel *et al., Science* 244:1281-1288, 1989), which cell or organisms are rendered transgenic by the introduction of the heterologous HRPC-related cDNA.

For expression in mammalian cells, the cDNA sequence may be ligated to heterologous promoters, such as the simian virus (SV) 40 promoter in the pSV2 vector (Mulligan and Berg, *Proc. Natl. Acad. Sci. USA* 78:2072-2076, 1981), and introduced into cells, such as monkey COS-1 cells (Gluzman, *Cell* 23:175-182, 1981), to achieve transient or long-term expression. The stable integration of the chimeric gene construct may be maintained in mammalian cells by biochemical selection, for example with neomycin (Southern and Berg, *J. Mol. Appl. Genet.* 1:327-341, 1982) or mycophenolic acid (Mulligan and Berg, *Proc. Natl. Acad. Sci. USA* 78:2072-2076, 1981).

DNA sequences can be manipulated with standard procedures such as restriction enzyme digestion, fill-in with DNA polymerase, deletion by exonuclease, extension by terminal deoxynucleotide transferase, ligation of synthetic or cloned DNA sequences, site-directed sequence-alteration via single-stranded bacteriophage intermediate or with the use of specific oligonucleotides in combination with PCR.

The cDNA sequence (or portions derived from it) or a mini gene (a cDNA with an intron and its own promoter) may be introduced into eukaryotic expression vectors by conventional techniques. These vectors are designed to permit the transcription of the cDNA in eukaryotic cells by providing regulatory sequences that initiate and enhance the transcription of the cDNA and ensure its proper splicing and polyadenylation. Vectors containing the promoter and enhancer regions of the SV40 or long terminal repeat (LTR) of the Rous Sarcoma virus and polyadenylation and splicing signal from SV40 are readily available (Mulligan *et al., Proc. Natl. Acad. Sci. USA* 78:1078-2076, 1981; Gorman *et al., Proc. Natl. Acad. Sci USA* 78:6777-6781, 1982). The level of expression of the cDNA can be manipulated with this type of vector, either by using promoters that have different activities (for example, the baculovirus pAC373 can express cDNAs at high levels in *S. frugiperda* cells (Summers and Smith, In *Genetically Altered Viruses and the Environment*, Fields *et al.* (Eds.) 22:319-328, CSHL Press, Cold Spring Harbor, New York, 1985) or by using vectors that contain promoters amenable to modulation, for example, the glucocorticoid-responsive promoter from the mouse mammary tumor virus (Lee *et al., Nature* 294:228, 1982). The expression of the cDNA can be monitored in the recipient cells 24 to 72 hours after introduction (transient expression).

In addition, some vectors contain selectable markers such as the *gpt* (Mulligan and Berg, *Proc. Natl. Acad. Sci. USA* 78:2072-2076, 1981) or *neo* (Southern and Berg, *J. Mol. Appl. Genet.* 1:327-341, 1982) bacterial genes. These selectable markers permit selection of transfected cells that exhibit stable, long-term expression of the vectors (and therefore the cDNA). The vectors can be maintained in the cells as episomal, freely replicating entities by using regulatory elements of viruses such as papilloma (Sarver *et al., Mol. Cell Biol.* 1:486, 1981) or Epstein-Barr (Sugden *et al., Mol.*

Cell Biol. 5:410, 1985). Alternatively, one can also produce cell lines that have integrated the vector into genomic DNA. Both of these types of cell lines produce the gene product on a continuous basis. One can also produce cell lines that have amplified the number of copies of the vector (and therefore of the cDNA as well) to create cell lines that can produce high levels of the gene product (Alt *et al.*, *J. Biol. Chem.* 253:1357, 1978).

The transfer of DNA into eukaryotic, in particular human or other mammalian cells, is now a conventional technique. The vectors are introduced into the recipient cells as pure DNA (transfection) by, for example, precipitation with calcium phosphate (Graham and vander Eb, *Virology* 52:466, 1973) or strontium phosphate (Brash *et al., Mol. Cell Biol.* 7:2013, 1987), electroporation (Neumann *et al., EMBO J* 1:841, 1982), lipofection (Felgner *et al., Proc. Natl. Acad. Sci USA* 84:7413, 1987), DEAE dextran (McCuthan *et al., J. Natl. Cancer Inst.* 41:351, 1968), microinjection (Mueller *et al., Cell* 15:579, 1978), protoplast fusion (Schafner, *Proc. Natl. Acad. Sci. USA* 77:2163-2167, 1980), or pellet guns (Klein *et al., Nature* 327:70, 1987). Alternatively, the cDNA, or fragments thereof, can be introduced by infection with virus vectors. Systems are developed that use, for example, retroviruses (Bernstein *et al., Gen. Engr'g* 7:235, 1985), adenoviruses (Ahmad *et al., J. Virol.* 57:267, 1986), or Herpes virus (Spaete *et al., Cell* 30:295, 1982). MB1 encoding sequences can also be delivered to target cells *in vitro* via non-infectious systems, for instance liposomes.

These eukaryotic expression systems can be used for studies of HRPC-related nucleic acids (such as those listed in Table 1) and mutant forms of these molecules, as well as HRPC-related proteins and mutant forms of these protein. Such uses include, for example, the identification of regulatory elements located in the 5' region of HRPC-related genes on genomic clones that can be isolated from human genomic DNA libraries. The eukaryotic expression systems may also be used to study the function of the normal HRPC-related proteins, specific portions of these proteins, or of naturally occurring or artificially produced mutant versions of HRPC-related proteins.

Using the above techniques, the expression vectors containing HRPC-related gene sequence or cDNA, or fragments or variants or mutants thereof, can be introduced into human cells, mammalian cells from other species or non-mammalian cells as desired. The choice of cell is determined by the purpose of the treatment. For example, monkey COS cells (Gluzman, *Cell* 23:175-182, 1981) that produce high levels of the SV40 T antigen and permit the replication of vectors containing the SV40 origin of replication may be used. Similarly, Chinese hamster ovary (CHO), mouse NIH 3T3 fibroblasts or human fibroblasts or lymphoblasts may be used.

The present disclosure thus encompasses recombinant vectors that comprise all or part of a HRPC-related gene or cDNA sequence, for expression in a suitable host. The HRPC-related nucleic acid sequence is operatively linked in the vector to an expression control sequence to form a recombinant DNA molecule, so that the HRPC-related polypeptide can be expressed. The expression control sequence may be selected from the group consisting of sequences that control the expression of genes of prokaryotic or eukaryotic cells and their viruses and combinations thereof. The

expression control sequence may be specifically selected from the group consisting of the *lac* system, the *trp* system, the *tac* system, the *trc* system, major operator and promoter regions of phage lambda, the control region of fd coat protein, the early and late promoters of SV40, promoters derived from polyoma, adenovirus, retrovirus, baculovirus and simian virus, the promoter for 3-phosphoglycerate kinase, the promoters of yeast acid phosphatase, the promoter of the yeast alpha-mating factors and combinations thereof.

The host cell, which may be transfected with the vector of this disclosure, may be selected from the group consisting of *E. coli, Pseudomonas, Bacillus subtilis, B. stearothermophilus* or other bacilli; other bacteria; yeast; fungi; insect; mouse or other animal; or plant hosts; or human tissue cells.

It is appreciated that for mutant or variant HRPC-related DNA sequences, similar systems are employed to express and produce the mutant product. In addition, fragments of a HRPC-related protein can be expressed essentially as detailed above. Such fragments include individual HRPC-related protein domains or sub-domains, as well as shorter fragments such as peptides. HRPC-related protein fragments having therapeutic properties may be expressed in this manner also.

EXAMPLE 8

Suppression of HRPC-related Gene Expression

A reduction of HRPC-related protein expression in a transgenic cell may be obtained by introducing into cells an antisense construct based on a HRPC-related protein encoding sequence, such as a cDNA or gene sequence or flanking regions thereof of any one of the proteins listed in Table 1, Table 4, or elsewhere herein. For antisense suppression, a nucleotide sequence encoding a HRPC-related protein, *e.g.* all or a portion of the cartilage glycoprotein-39 (CHI3L1), S-100P PROTEIN (S100P), CX3C chemokine/fractalkine (SCYD1), adenylate kinase 1 (AK1), forkhead transcription factor HFH-4 (HFH-4) (FKHL13), UDP glucuronosyltransferase precursor (UGT2B15), Pleiotrophin (heparin binding growth factor 8) (PTN), heat shock 27kD protein 2/Alpha-B-crystallin (HSP27), Proteasome (prosome, macropain) subunit, beta type, 5 (PSMB5), Inhibitor of NFKB (NFKBIA), interferon-induced 17 kD protein (ISG15), MAP kinase activated protein kinase 2 (MAPKAPK2), signal transduction protein (SH3 containing) (EFS2), hkf-1 Zinc finger protein (ZFP103), chromosome condensation 1 (CHC1), CDP-diacylglycerol synthase (CDS1), gap junction protein, alpha 1, 43kD (connexin 43) (GJA1), cyclin D1 (CCND1), Inhibitor of DNA binding 3, helix-loop-helix protein (ID3), H1 histone family, member2 (H1F2), Cytochrome B561 (CYB561), Cathepsin H (CTSH), calcineurin alpha (PPP3CA), 54 kDa progesterone receptor-associated immunophilin (FKBP5), translocation protein 1 (TLOC1), Clusterin (complement lysis inhibitor; testosterone-repressed prostate message 2; apolipoprotein J) (CLU), Pulmonary surfactant-associated protein A (SFTPA1), protease inhibitor 12 (PI12; neuroserpin) (PI12), Thrombospondin 1 (THBS1), Ribophorin I (RPN1), A disintegrin-like and metalloprotease (reprolysin type) with thrombospondin type 1 motif, 1 (ADAMTS1), Collagen, type IV, alpha 5 (Alport syndrome) (COL4A5), LIM domain

only 4 / breast tumor autoantigen (LMO4), bumetanide-sensitive Na-K-Cl cotransporter (NKCC1) (SLC12A2), Fibronectin (FN1), Crystallin Mu (CRYM) or "upregulated by 1,25-dihydroxyvitamin D-3" (VDUP1) cDNA or gene, is arranged in reverse orientation relative to the promoter sequence in the transformation vector. Other aspects of the vector may be chosen as for any other expression vector (see, *e.g.*, Example 7).

The introduced sequence need not be a full-length human HRPC-related cDNA or gene, and need not be exactly homologous to the equivalent sequence found in the cell type to be transformed. Generally, however, where the introduced sequence is of shorter length, a higher degree of homology to the HRPC-related sequence likely will be needed for effective antisense suppression. The introduced antisense sequence in the vector may be at least 30 nucleotides in length, and improved antisense suppression will typically be observed as the length of the antisense sequence increases. The length of the antisense sequence in the vector advantageously may be greater than 100 nucleotides.

Although the exact mechanism by which antisense RNA molecules interfere with gene expression has not been elucidated, it is believed that antisense RNA molecules bind to the endogenous mRNA molecules and thereby inhibit translation of the endogenous mRNA.

Suppression of endogenous HRPC-related gene expression can also be achieved using ribozymes. Ribozymes are synthetic RNA molecules that possess highly specific endoribonuclease activity. The production and use of ribozymes are disclosed in U.S. Patent No. 4,987,071 to Cech and U.S. Patent No. 5,543,508 to Haselhoff. The inclusion of ribozyme sequences within antisense RNAs may be used to confer RNA cleaving activity on the antisense RNA, such that endogenous mRNA molecules that bind to the antisense RNA are cleaved, which in turn leads to an enhanced antisense inhibition of endogenous gene expression.

In addition, dominant negative mutant forms of the disclosed HRPC-related sequences may be used to block endogenous activity of the corresponding gene products.

Suppression can also be achieved using small inhibitory RNA molecules (siRNAs) (see, for instance, Caplen *et al.*, *Proc. Natl. Acad. Sci.* 98(17):9742-9747, 2001, and Elbashir *et al.*, *Nature* 411:494-498, 2001). Thus, this disclosure also encompasses siRNAs that correspond to an HRPC-related nucleic acid, which siRNA is capable of suppressing the expression or function of its cognate (target) HRPC-related protein. Also encompassed are methods of suppressing the expression or activity of an HRPC-related molecule using an siRNA.

Suppression of expression of an HRPC-related gene can be used, for instance, to treat, reduce, or prevent cell proliferative and other disorders caused by over-expression or unregulated expression of the corresponding HRPC-related gene. In particular, suppression of expression of sequences disclosed herein as being up-regulated in hormone-refractory prostate cancer can be used to treat, reduce, or prevent progression to hormone-refractory prostate cancer.

EXAMPLE 9

Production of Protein Specific Binding Agents

Monoclonal or polyclonal antibodies may be produced to any of the disclosed HRPC-related proteins, or mutant forms of these proteins. Optimally, antibodies raised against these proteins, or peptides from within such proteins, would specifically detect the protein or peptide with which the antibodies are generated. That is, an antibody generated to the S100P protein (or another specified protein) or a fragment thereof would recognize and bind that protein and would not substantially recognize or bind to other proteins found in human cells.

The determination that an antibody specifically detects a designated protein (*e.g.*, a HRPC-related protein as disclosed herein) can be made by any one of a number of standard immunoassay methods; for instance, the Western blotting technique (Sambrook *et al.*, In *Molecular Cloning: A Laboratory Manual*, CSHL, New York, 1989). To determine that a given antibody preparation (such as one produced in a mouse) specifically detects a designated protein by Western blotting, total cellular proteins are extracted from cells (for example, human prostate) and electrophoresed on a sodium dodecyl sulfate-polyacrylamide gel. The proteins are then transferred to a membrane (for example, nitrocellulose) by Western blotting, and the antibody preparation is incubated with the membrane. After washing the membrane to remove non-specifically bound antibodies, the presence of specifically bound antibodies is detected by the use of an anti-mouse antibody conjugated to an enzyme such as alkaline phosphatase. Application of an alkaline phosphatase substrate 5-bromo-4-chloro-3-indolyl phosphate/nitro blue tetrazolium results in the production of a dense blue compound by immunolocalized alkaline phosphatase. Antibodies that specifically detect the designated protein will, by this technique, be shown to bind to the designated protein band (which will be localized at a given position on the gel determined by its molecular weight). Non-specific binding of the antibody to other proteins may occur and may be detectable as a weak signal on the Western blot. The non-specific nature of this binding will be recognized by one skilled in the art by the weak signal obtained on the Western blot relative to the strong primary signal arising from the specific antibody-protein binding.

Substantially pure HRPC-related protein or protein fragment (peptide) suitable for use as an immunogen may be isolated from transfected or transformed cells, as described above. Concentration of protein or peptide in the final preparation is adjusted, for example, by concentration on an Amicon filter device, to the level of a few micrograms per milliliter. Monoclonal or polyclonal antibody to the protein can then be prepared as follows:

A. Monoclonal Antibody Production by Hybridoma Fusion

Monoclonal antibody to epitopes of a designated protein (such as a HRPC-related protein, including any one of those listed in Table 1) identified and isolated as described can be prepared from murine hybridomas according to the classical method of Kohler and Milstein (*Nature* 256:495-497, 1975) or derivative methods thereof. Briefly, a mouse is repetitively inoculated with a few

micrograms of the selected protein over a period of a few weeks. The mouse is then sacrificed, and the antibody-producing cells of the spleen isolated. The spleen cells are fused by means of polyethylene glycol with mouse myeloma cells, and the excess un-fused cells destroyed by growth of the system on selective media comprising aminopterin (HAT media). The successfully fused cells are diluted and aliquots of the dilution placed in wells of a microtiter plate where growth of the culture is continued. Antibody-producing clones are identified by detection of antibody in the supernatant fluid of the wells by immunoassay procedures, such as ELISA, as originally described by Engvall (*Meth. Enzymol.* 70:419-439, 1980), and derivative methods thereof. Selected positive clones can be expanded and their monoclonal antibody product harvested for use. Detailed procedures for monoclonal antibody production are described in Harlow and Lane (*Antibodies, A Laboratory Manual*, CSHL, New York, 1988).

B. Polyclonal Antibody Production by Immunization

Polyclonal antiserum containing antibodies to heterogeneous epitopes of a single protein can be prepared by immunizing suitable animals with the expressed protein (Example 7), which can be unmodified or modified to enhance immunogenicity. Effective polyclonal antibody production is affected by many factors related both to the antigen and the host species. For example, small molecules tend to be less immunogenic than others and may require the use of carriers and adjuvant. Also, host animals vary in response to site of inoculations and dose, with either inadequate or excessive doses of antigen resulting in low titer antisera. Small doses (ng level) of antigen administered at multiple intradermal sites appear to be most reliable. An effective immunization protocol for rabbits can be found in Vaitukaitis *et al.* (*J. Clin. Endocrinol. Metab.* 33:988-991, 1971).

Booster injections can be given at regular intervals, and antiserum harvested when antibody titer thereof, as determined semi-quantitatively, for example, by double immunodiffusion in agar against known concentrations of the antigen, begins to fall. See, for example, Ouchterlony *et al.* (In *Handbook of Experimental Immunology*, Wier (ed.) chapter 19. Blackwell, 1973). Plateau concentration of antibody is usually in the range of about 0.1 to 0.2 mg/ml of serum (about 12 μM). Affinity of the antisera for the antigen is determined by preparing competitive binding curves, as described, for example, by Fisher (*Manual of Clinical Immunology*, Ch. 42, 1980).

C. Antibodies Raised against Synthetic Peptides

A third approach to raising antibodies against the subject HRPC-related proteins or peptides is to use one or more synthetic peptides synthesized on a commercially available peptide synthesizer based upon the predicted amino acid sequence of the desired HRPC-related protein or peptide.

D. Antibodies Raised by Injection of Protein Encoding Sequence

Antibodies also may be raised against proteins and peptides related to HRPC as described herein by subcutaneous injection of a DNA vector that expresses the desired HRPC-related protein, or a fragment thereof, into laboratory animals, such as mice. Delivery of the recombinant vector into the animals may be achieved using a hand-held form of the Biolistic system (Sanford *et al.*, *Particulate Sci. Technol.* 5:27-37, 1987) as described by Tang *et al.* (*Nature* 356:152-154, 1992).

Expression vectors suitable for this purpose may include those that express the HRPC-related sequence under the transcriptional control of either the human β-actin promoter or the cytomegalovirus (CMV) promoter.

Antibody preparations prepared according to these protocols are useful in quantitative immunoassays which determine concentrations of antigen-bearing substances in biological samples; they also can be used semi-quantitatively or qualitatively to identify the presence of antigen in a biological sample; or for immunolocalization of the corresponding HRPC-related protein.

For administration to human patients, antibodies, *e.g.*, HRPC-related protein specific monoclonal antibodies (such as antibodies to the proteins encoded by the encoding sequences referred to in Table 1), can be humanized by methods known in the art. Antibodies with a desired binding specificity can be commercially humanized (Scotgene, Scotland, UK; Oxford Molecular, Palo Alto, CA). Alternatively, human antibodies can be produced. Methods for producing human antibodies are known in the art; see, for instance, Canevari *et al.*, *Int J Biol Markers* 8:147-150, 1993 and Green, *J Immunol Methods* 231:11-23, 1999, for instance.

EXAMPLE 10

Nucleic Acid-Based Analysis

The HRPC-related nucleic acid molecules provided herein can be used in methods of genetic testing for neoplasms (*e.g.*, prostate or other cancers) or predisposition to neoplasms owing to HRPC-related nucleic acid molecule deletion, genomic amplification or mutation, or over- or under-expression in comparison to a control or baseline. For such procedures, a biological sample of the subject, which biological sample contains either DNA or RNA derived from the subject, is assayed for a mutated, amplified or deleted HRPC-related nucleic acid molecule, or for over- or under expression of a HRPC-related nucleic acid molecule. Suitable biological samples include samples containing genomic DNA or RNA (including mRNA), obtained from body cells of a subject, such as those present in peripheral blood, urine, saliva, tissue biopsy, surgical specimen, amniocentesis samples and autopsy material.

The detection in the biological sample of a mutant HRPC-related nucleic acid molecule, a mutant HRPC-related RNA, an amplified or homozygously or heterozygously deleted HRPC-related nucleic acid molecule, or over- or under-expression of a HRPC-related nucleic acid molecule, may be performed by a number of methodologies.

A. Detection of Unknown Mutations:

Unknown mutations in HRPC-related nucleic acid molecules can be identified through polymerase chain reaction amplification of reverse transcribed RNA (RT-PCR) or DNA isolated from breast or other tissue, followed by direct DNA sequence determination of the products; single-strand conformational polymorphism analysis (SSCP) (for instance, see Hongyo *et al.*, *Nucleic Acids Res.* 21:3637-3642, 1993); chemical cleavage (including HOT cleavage) (Bateman *et al.*, *Am. J. Med. Genet.* 45:233-240, 1993; reviewed in Ellis *et al.*, *Hum. Mutat.* 11:345-353, 1998); denaturing

gradient gel electrophoresis (DGGE), ligation amplification mismatch protection (LAMP); or enzymatic mutation scanning (Taylor and Deeble, *Genet. Anal.* 14:181-186, 1999), followed by direct sequencing of amplicons with putative sequence variations.

B. Detection of Known Mutations:

The detection of specific known DNA mutations in HRPC-related nucleic acid molecules may be achieved by methods such as hybridization using allele specific oligonucleotides (ASOs) (Wallace *et al.*, CSHL Symp. Quant. Biol. 51:257-261, 1986), direct DNA sequencing (Church and Gilbert, *Proc. Natl. Acad. Sci. USA* 81:1991-1995, 1988), the use of restriction enzymes (Flavell *et al., Cell* 15:25, 1978; Geever *et al.*, 1981), discrimination on the basis of electrophoretic mobility in gels with denaturing reagent (Myers and Maniatis, Cold Spring Harbor Symp. Quant. Biol. 51:275-284, 1986), RNase protection (Myers *et al., Science* 230:1242, 1985), chemical cleavage (Cotton *et al., Proc. Natl. Acad. Sci. USA* 85:4397-4401, 1985), and the ligase-mediated detection procedure (Landegren *et al., Science* 241:1077, 1988). Oligonucleotides specific to normal or mutant MB1 sequences can be chemically synthesized using commercially available machines. These oligonucleotides can then be labeled radioactively with isotopes (such as ^{32}P) or non-radioactively, with tags such as biotin (Ward and Langer *et al., Proc. Natl. Acad. Sci. USA* 78:6633-6657, 1981), and hybridized to individual DNA samples immobilized on membranes or other solid supports by dot-blot or transfer from gels after electrophoresis. These specific sequences are visualized by methods such as autoradiography or fluorometric (Landegren *et al., Science* 242:229-237, 1989) or colorimetric reactions (Gebeyehu *et al.*, *Nucleic Acids Res.* 15:4513-4534, 1987). Using an ASO specific for a normal allele, the absence of hybridization would indicate a mutation in the particular region of the gene, or deleted MB1 gene. In contrast, if an ASO specific for a mutant allele hybridizes to a clinical sample then that would indicate the presence of a mutation in the region defined by the ASO.

C. Detection of Genomic Amplification or Deletion

Gene dosage (copy number) can be important in neoplasms; it is therefore advantageous to determine the number of copies of HRPC-related nucleic acids in biological samples of a subject, *e.g.*, serum or prostate samples. Probes generated from the disclosed encoding sequence of in HRPC-related nucleic acid molecules can be used to investigate and measure genomic dosage of the corresponding HRPC-related genomic sequence.

Appropriate techniques for measuring gene dosage are known in the art; see for instance, US Patent No. 5,569,753 ("Cancer Detection Probes") and Pinkel *et al.* (*Nat. Genet.* 20:207-211, 1998) ("High Resolution Analysis of DNA Copy Number Variation using Comparative Genomic Hybridization to Microarrays").

Determination of gene copy number in cells of a patient-derived sample using other techniques is known in the art. For example, amplification of a HRPC-related nucleic acid sequence in cancer-derived cell lines as well as uncultured prostate cancer or other cells can be carried out using bicolor FISH analysis. By way of example, interphase FISH analysis of breast cancer cell lines

can be carried out as previously described (Barlund *et al.*, *Genes Chromo. Cancer* 20:372-376, 1997). The hybridizations can be evaluated using a Zeiss fluorescence microscope.

For tissue microarrays, the FISH can be performed as described in Kononen *et al.*, *Nat. Med.* 4:844-847, 1998. Briefly, consecutive sections of the array are deparaffinized, dehydrated in ethanol, denatured at 74° C for 5 minutes in 70% formamide/2 x SSC, and hybridized with test and reference probes. The specimens containing tight clusters of signals or >3-fold increase in the number of test probe as compared to chromosome 17 centromere in at least 10% of the tumor cells may be considered as amplified. Microarrays can be constructed as described in WO99/44063A2 and WO99/44062A1.

C. Detection of mRNA Expression Levels

Over- or under-expression of a HRPC-related molecule can also be detected by measuring the cellular level of HRPC-related nucleic acid molecule-specific mRNA. mRNA can be measured using techniques well known in the art, including for instance Northern analysis, RT-PCR and mRNA *in situ* hybridization. Details of such procedures can be found, for instance, in Examples 1 and 3.

The nucleic acid-based diagnostic methods of this disclosure are predictive of proliferation, metastatic potential, cancer progression, and response to treatment in patients suffering from prostate carcinomas including hormone-refractory prostate carcinomas, and other solid tumors, carcinomas, sarcomas, and cancers. Cells of any tumors that demonstrate abnormal levels (*e.g.*, through genomic amplification, deletion, mutation, or other over- or under-expression) of nucleotide sequences that share homology with the HRPC-related nucleic acids disclosed herein are aggressive tumor cells, and result in decreased survival, increased metastasis, increased rates of clinical recurrence (such as recurrence after hormone ablation therapy), and overall worsened prognosis.

EXAMPLE 11

Protein-Based Analysis

An alternative method of diagnosing, staging, detecting, or predicting hormone-related prostate cancer is to quantitate the level of one or more HRPC-related proteins in a subject, for instance in the cells of the subject. This diagnostic tool is useful for detecting reduced or increased levels of HRPC-related proteins. Localization and/or coordinated expression (temporally or spatially) of HRPC-related proteins can also be examined using well known techniques. The determination of reduced or increased HRPC-related protein levels, in comparison to such expression in a normal subject (*e.g.*, a subject not having hormone-related prostate cancer or not having a predisposition developing this condition, disease or disorder, would be an alternative or supplemental approach to the direct determination of HRPC-related nucleic acid levels by the methods outlined above and equivalents. The availability of antibodies specific to specific HRPC-related protein(s) will facilitate the detection and quantitation of cellular HRPC-related protein(s) by one of a number of immunoassay methods which are well known in the art and are presented in Harlow and Lane

(*Antibodies, A Laboratory Manual*, CSHL, New York, 1988). Methods of constructing such antibodies are discussed above, in Example 9.

Any standard immunoassay format (*e.g.*, ELISA, Western blot, or RIA assay) can be used to measure HRPC-related polypeptide or protein levels; comparison is to wild-type (normal) HRPC-related protein levels, and a difference in HRPC-related polypeptide levels is indicative of an abnormal biological condition such as neoplasia. Whether the key difference is an increase or a decrease is dependent on the specific HRPC-related protein under examination, as discussed herein. Immunohistochemical techniques may also be utilized for HRPC-related polypeptide or protein detection and quantification. For example, a tissue sample may be obtained from a subject, and a section stained for the presence of a HRPC-related protein using the appropriate HRPC-related protein specific binding agent and any standard detection system (*e.g.*, one which includes a secondary antibody conjugated to horseradish peroxidase). General guidance regarding such techniques can be found in, *e.g.*, Bancroft and Stevens (*Theory and Practice of Histological Techniques*, Churchill Livingstone, 1982) and Ausubel *et al.* (*Current Protocols in Molecular Biology*, John Wiley & Sons, New York, 1998).

For the purposes of quantitating a HRPC-related protein, a biological sample of the subject, which sample includes cellular proteins, is required. Such a biological sample may be obtained from body cells, such as those present in peripheral blood, urine, saliva, tissue biopsy, amniocentesis samples, surgical specimens and autopsy material, particularly breast cells. Quantitation of a HRPC-related protein can be achieved by immunoassay and the amount compared to levels of the protein found in healthy cells. A significant difference (either increase or decrease) in the amount of HRPC-related protein in the cells of a subject compared to the amount of the same HRPC-related protein found in normal human cells is usually about a 30% or greater difference. Substantial under- or over-expression of one or more HRPC-related protein(s), may be indicative of neoplasia or a predilection to neoplasia or metastasis, and especially hormone-refractory prostate cancer.

The protein-based diagnostic methods as described herein are predictive of proliferation, metastatic potential, cancer progression, and response to treatment in patients suffering from prostate carcinomas including hormone-refractory prostate carcinomas, and other solid tumors, carcinomas, sarcomas, and cancers. Cells of any tumors that demonstrate abnormal levels (*e.g.*, through genomic amplification, deletion, mutation, or other over- or under-expression) of nucleotide sequences that share homology with the HRPC-related nucleic acids disclosed herein are aggressive tumor cells, and result in decreased survival, increased metastasis, increased rates of clinical recurrence (such as recurrence after hormone ablation therapy), and overall worsened prognosis.

EXAMPLE 12:

Gene Therapy

Gene therapy approaches for combating neoplasia (particularly prostate cancer, including hormone-refractory prostate cancer) in subjects are made possible by the present disclosure.

Retroviruses have been considered a preferred vector for experiments in gene therapy, with a high efficiency of infection and stable integration and expression (Orkin *et al., Prog. Med. Genet.* 7:130-142, 1988). A full-length HRPC-related gene or cDNA can be cloned into a retroviral vector and driven from either its endogenous promoter or from the retroviral LTR (long terminal repeat). Other viral transfection systems may also be utilized for this type of approach, including adenovirus, adeno-associated virus (AAV) (McLaughlin *et al., J. Virol.* 62:1963-1973, 1988), *Vaccinia* virus (Moss *et al., Annu. Rev. Immunol.* 5:305-324, 1987), Bovine Papilloma virus (Rasmussen *et al., Methods Enzymol.* 139:642-654, 1987) or members of the herpesvirus group such as Epstein-Barr virus (Margolskee *et al., Mol. Cell. Biol.* 8:2837-2847, 1988).

Recent developments in gene therapy techniques include the use of RNA-DNA hybrid oligonucleotides, as described by Cole-Strauss, *et al.* (*Science* 273:1386-1389, 1996). This technique may allow for site-specific integration of cloned sequences, thereby permitting accurately targeted gene replacement.

In addition to delivery of HRPC-related protein encoding sequences to cells using viral vectors, it is possible to use non-infectious methods of delivery. For instance, lipidic and liposome-mediated gene delivery has recently been used successfully for transfection with various genes (for reviews, see Templeton and Lasic, *Mol. Biotechnol.* 11:175-180, 1999; Lee and Huang, *Crit. Rev. Ther. Drug Carrier Syst.* 14:173-206; and Cooper, *Semin. Oncol.* 23:172-187, 1996). For instance, cationic liposomes have been analyzed for their ability to transfect monocytic leukemia cells, and shown to be a viable alternative to using viral vectors (de Lima *et al.*, *Mol. Membr. Biol.* 16:103-109, 1999). Such cationic liposomes can also be targeted to specific cells through the inclusion of, for instance, monoclonal antibodies or other appropriate targeting ligands (Kao *et al.*, *Cancer Gene Ther.* 3:250-256, 1996).

To reduce the level of HRPC-related gene expression, gene therapy can be carried out using antisense or other suppressive constructs, the construction of which is discussed above (Example 8).

EXAMPLE 13

Kits

Kits are provided to determine the level (or relative level) of expression of one or more species of HRPC-related mRNA (*i.e.*, kits containing probes) or one or more HRPC-related protein (*i.e.*, kits containing antibodies or other HRPC-related protein specific binding agents). Kits are also provided that contain the necessary reagents for determining gene copy number (genomic amplification or deletion), such as probes or primers specific for a HRPC-related nucleic acid sequence. These kits can each include instructions, for instance instructions that provide calibration curves or charts to compare with the determined (*e.g.*, experimentally measured) values.

A. Kits for Detection of HRPC-related Genomic Amplification or Deletion

The nucleotide sequence of HRPC-related nucleic acid molecules disclosed herein, and fragments thereof, can be supplied in the form of a kit for use in detection of HRPC-related genomic amplification/deletion and/or diagnosis of progression to or predilection to progress to hormone-refractory prostate cancer. In such a kit, an appropriate amount of one or more oligonucleotide primer specific for an HRPC-related-sequence is provided in one or more containers. The oligonucleotide primers may be provided suspended in an aqueous solution or as a freeze-dried or lyophilized powder, for instance. The container(s) in which the oligonucleotide(s) are supplied can be any conventional container that is capable of holding the supplied form, for instance, microfuge tubes, ampoules, or bottles. In some applications, pairs of primers may be provided in pre-measured single use amounts in individual, typically disposable, tubes, or equivalent containers. With such an arrangement, the sample to be tested for the presence of HRPC-related genomic amplification/deletion can be added to the individual tubes and *in vitro* amplification carried out directly.

The amount of each oligonucleotide primer supplied in the kit can be any amount, depending for instance on the market to which the product is directed. For instance, if the kit is adapted for research or clinical use, the amount of each oligonucleotide primer provided likely would be an amount sufficient to prime several *in vitro* amplification reactions. Those of ordinary skill in the art know the amount of oligonucleotide primer that is appropriate for use in a single amplification reaction. General guidelines may for instance be found in Innis *et al.* (*PCR Protocols, A Guide to Methods and Applications*, Academic Press, Inc., San Diego, CA, 1990), Sambrook *et al.* (In *Molecular Cloning: A Laboratory Manual,* Cold Spring Harbor, New York, 1989), and Ausubel *et al.* (In *Current Protocols in Molecular Biology*, John Wiley & Sons, New York, 1998).

A kit may include more than two primers, in order to facilitate the *in vitro* amplification of HRPC-related genomic sequences, for instance a HRPC-related nucleic acid listed in Table 1, or the 5' or 3' flanking region thereof.

In some embodiments, kits may also include the reagents necessary to carry out *in vitro* amplification reactions, including, for instance, DNA sample preparation reagents, appropriate buffers (*e.g.*, polymerase buffer), salts (*e.g.*, magnesium chloride), and deoxyribonucleotides (dNTPs). Written instructions may also be included.

Kits may in addition include either labeled or unlabeled oligonucleotide probes for use in detection of the *in vitro* amplified sequences. The appropriate sequences for such a probe will be any sequence that falls between the annealing sites of two provided oligonucleotide primers, such that the sequence the probe is complementary to is amplified during the *in vitro* amplification reaction (if it is present in the sample).

It may also be advantageous to provided in the kit one or more control sequences for use in the *in vitro* amplification reactions. The design of appropriate positive control sequences is well known to one of ordinary skill in the appropriate art.

B. Kits for Detection of mRNA Expression

Kits similar to those disclosed above for the detection of HRPC-related genomic amplification/deletion can be used to detect HRPC-related mRNA expression levels (including over- or under-expression, in comparison to the expression level in a control sample). Such kits include an appropriate amount of one or more of the oligonucleotide primers for use in, for instance, reverse transcription PCR reactions, similarly to those provided above, with art-obvious modifications for use with RNA.

In some embodiments, kits for detection of HRPC-related mRNA expression may also include reagents necessary to carry out RT-PCR or other *in vitro* amplification reactions, including, for instance, RNA sample preparation reagents (including *e.g.*, an RNAse inhibitor), appropriate buffers (*e.g.*, polymerase buffer), salts (*e.g.*, magnesium chloride), and deoxyribonucleotides (dNTPs). Written instructions may also be included.

Kits may in addition include either labeled or unlabeled oligonucleotide probes for use in detection of an *in vitro* amplified target sequence. The appropriate sequences for such a probe will be any sequence that falls between the annealing sites of the two provided oligonucleotide primers, such that the sequence the probe is complementary to is amplified during the PCR reaction.

It may also be advantageous to provided in the kit one or more control sequences for use in the *in vitro* amplification reactions. The design of appropriate positive control sequences is well known to one of ordinary skill in the appropriate art.

Alternatively, kits may be provided with the necessary reagents to carry out quantitative or semi-quantitative Northern analysis of HRPC-related mRNA. Such kits include, for instance, at least one HRPC-related sequence-specific oligonucleotide for use as a probe. This oligonucleotide may be labeled in any conventional way, including with a selected radioactive isotope, enzyme substrate, co-factor, ligand, chemiluminescent or fluorescent agent, hapten, or enzyme.

C. Kits For Detection of HRPC-linked Protein or Peptide Expression

Kits for the detection of HRPC-linked protein expression, for instance abnormal (over or under) expression of a protein encoded for by a nucleic acid molecule listed in Table 1, are also encompassed herein. Such kits will include at least one target (HRPC-linked) protein (*e.g.*, cartilage glycoprotein-39 (CHI3L1), S-100P PROTEIN (S100P), CX3C chemokine/fractalkine (SCYD1), adenylate kinase 1 (AK1), forkhead transcription factor HFH-4 (HFH-4) (FKHL13), UDP glucuronosyltransferase precursor (UGT2B15), Pleiotrophin (heparin binding growth factor 8) (PTN), heat shock 27kD protein 2/Alpha-B-crystallin (HSP27), Proteasome (prosome, macropain) subunit, beta type, 5 (PSMB5), Inhibitor of NFKB (NFKBIA), interferon-induced 17 kD protein (ISG15), MAP kinase activated protein kinase 2 (MAPKAPK2), signal transduction protein (SH3

containing) (EFS2), hkf-1 Zinc finger protein (ZFP103), chromosome condensation 1 (CHC1), CDP-diacylglycerol synthase (CDS1), gap junction protein, alpha 1, 43kD (connexin 43) (GJA1), cyclin D1 (CCND1), Inhibitor of DNA binding 3, helix-loop-helix protein (ID3), H1 histone family, member2 (H1F2), Cytochrome B561 (CYB561), Cathepsin H (CTSH), calcineurin alpha (PPP3CA), 54 kDa progesterone receptor-associated immunophilin (FKBP5), translocation protein 1 (TLOC1), Clusterin (complement lysis inhibitor; testosterone-repressed prostate message 2; apolipoprotein J) (CLU), Pulmonary surfactant-associated protein A (SFTPA1), protease inhibitor 12 (PI12; neuroserpin) (PI12), Thrombospondin 1 (THBS1), Ribophorin I (RPN1), A disintegrin-like and metalloprotease (reprolysin type) with thrombospondin type 1 motif, 1 (ADAMTS1), Collagen, type IV, alpha 5 (Alport syndrome) (COL4A5), LIM domain only 4 / breast tumor autoantigen (LMO4), bumetanide-sensitive Na-K-Cl cotransporter (NKCC1) (SLC12A2), Fibronectin (FN1), or Crystallin Mu (CRYM)) specific binding agent (*e.g.*, a polyclonal or monoclonal antibody or antibody fragment), and may include at least one control. The HRPC-linked protein specific binding agent and control may be contained in separate containers. The kits may also include a means for detecting HRPC-related protein:agent complexes, for instance the agent may be detectably labeled. If the detectable agent is not labeled, it may be detected by second antibodies or protein A, for example, either of both of which also may be provided in some kits in one or more separate containers. Such techniques are well known.

Additional components in some kits include instructions for carrying out the assay. Instructions will allow the tester to determine whether HRPC-linked expression levels are elevated or reduced in comparison to a control sample. Reaction vessels and auxiliary reagents such as chromogens, buffers, enzymes, etc. may also be included in the kits.

EXAMPLE 14

Identification of Therapeutic Compounds

The HRPC-related molecules disclosed herein, and more particularly the linkage of these molecules to cancer and cancer progression, can be used to identify compounds that are useful in treating, reducing, or preventing prostate cancer or development or progression of prostate cancer. These molecules can be used alone or in combination, for instance in sets of two or more that are linked to cancer or cancer progression.

By way of example, a test compound is applied to a cell, for instance a test cell, and at least one HRPC-related molecule level and/or activity in the cell is measured and compared to the equivalent measurement from a test cell (or from the same cell prior to application of the test compound). If application of the compound alters the level and/or activity of a HRPC-related molecule (for instance by increasing or decreasing that level), then that compound is selected as a likely candidate for further characterization. In particular examples, a test agent that opposes or inhibits an HRPC-related change is selected for further study, for example by exposing the agent to a hormone refractory prostate cancer cell *in vitro*, to determine whether *in vitro* growth is inhibited.

Such identified compounds may be useful in treating, reducing, or preventing prostate cancer or development or progression of prostate cancer. In particular embodiments, the compound isolated will inhibit or inactivate a HRPC-related molecule, for instance those represented by the nucleic acids listed in Table 1.

Methods for identifying such compounds optionally can include the generation of a HRPC-related gene expression profile, as described herein. Control gene expression profiles useful for comparison in such methods may be constructed from normal prostate tissue, primary prostate cancer tissue, prostate cancer tissue responding to androgen ablation therapy, and/or a hormone refractory prostate cancer tissue.

By way of specific example, rapamycin has been herein identified as a compound that influences the levels of HRPC-related molecules, in particular certain of the nucleic acid molecules listed in for instance Table 1 (as discussed in more detail above). With the provision herein of this identification, the use of rapamycin as a treatment for HRPC is now enabled, as is the use of rapamycin derivatives or rapamycin-like compounds. It is believed that rapamycin can be used on its own as such a treatment, or can be used in combination with known or newly identified treatments for HRPC.

EXAMPLE 15

Gene Expression Profiles (Fingerprints)

With the provision herein of methods for determining molecules that are linked to HRPC, and the provision of a large collection of such HRPC-linked molecules (as represented for instance by those listed in Table 1), gene expression profiles that provide information on the prostate cancer-state of a subject are now enabled.

HRPC-related expression profiles comprise the distinct and identifiable pattern of expression (or level) of sets of HRPC-related genes, for instance a pattern of high and low expression of a defined set of genes, or molecules that can be correlated to such genes, such as mRNA levels or protein levels or activities. The set of molecules in a particular profile will usually include at least one that is represented by (or correlated to) the following Image ID Clones: 781047, 785778, 842968, 769921, 898286, 204214, 814701, 435076, 531319, 415089, 898062, 453107, 785707, 795936, 700792, 34778, 46182, 769921, 783697, 451907, 711768, 416833, 810711, 789204, 789182, 725454, 951142, 49352, 273546, 46717, 855487, 41117, 26578, 684655, 45233, 814117, 810552, 739511, 283315, and 897774. In other examples of HRPC-related gene expression profiles, more than one molecule corresponding to the Image ID Clones listed in Table 1 are included in the profile. By way of example, any subset of the molecules listed in Table 1 (or corresponding to the molecules in this list) may be included in a single gene expression profile. Specific examples of such subsets include those molecules that show an increasing expression profile during prostate cancer progression, those that show a decreasing expression profile, those that are most highly correlated to a particular stage of prostate cancer progression, and so forth. Alternatively, gene expression profiles may be further

broken down by the manner of molecules included in the profile. Thus, certain examples of profiles may include a specific class of HRPC-related molecules, such as those molecules involved in cell cycle control.

Particular profiles are specific for a particular stage of normal tissue (*e.g.*, prostate tissue) growth or disease progression (*e.g.*, progression of prostate cancer). Thus, gene expression profiles can be established for a pre-prostate cancer tissue (*i.e.*, normal prostate tissue), a primary prostate cancer tissue, a prostate cancer tissue responding to androgen ablation therapy, and a hormone refractory prostate cancer tissue. Each of these profiles includes information on the expression level of at least one, but usually two or more, genes that are linked to prostate cancer (*e.g.*, HRPC-related genes). Such information can include relative as well as absolute expression levels of specific genes. Likewise, the value measured may be the relative or absolute level of protein expression, which can be correlated with a "gene expression level." Results from the gene expression profiles of an individual subject are often viewed in the context of a test sample compared to a baseline or control sample fingerprint.

The levels of molecules that make up a gene expression profile can be measured in any of various known ways, which may be specific for the type of molecule being measured. Thus, nucleic acid levels (such as direct gene expression levels, such as the level of mRNA expression) can be measured using specific nucleic acid hybridization reactions. Protein levels may be measured using standard protein assays, using immunologic-based assays (such as ELISAs and related techniques), or using activity assays, for instance. Examples for measuring nucleic acid and protein levels are provided herein; other methods are well known to those of ordinary skill in the art.

Examples of HRPC-related gene expression profiles can be in array format, such as a nucleotide (*e.g.*, polynucleotide) or protein array or microarray. The use of arrays to determine the presence and/or level of a collection of biological macromolecules is now well known (see, for example, methods described in published PCT application number US99/06860, describing hyproxia-related gene expression arrays). In array-based measurement methods, an array may be contacted with polynucleotides (in the case of a nucleic acid-based array) or polypeptides (in the case of a protein-based array) from a sample from a subject. The amount and/or position of binding of the subject's polynucleotides or polypeptides then can be determined, for instance to produce a gene expression profile for that subject. Such gene expression profile can be compared to another gene expression profile, for instance a control gene expression profile from a subject having a known prostate-related condition. Optionally, the subject's gene expression profile can be correlated with one or more appropriate treatments, which may be correlated with a control (or set of control) expression profiles for stages of prostate cancer progression, for instance.

This disclosure provides the identification of HRPC-related molecules that exhibit alterations in expression during development of refractory prostate cancer, and expression fingerprints (profiles) specific for prostate cancer stages. It further provides methods of using these

identified nucleic acid molecules, and proteins encoded thereby, and expression fingerprints or profiles, to predict and/or diagnose hormone-refractory prostate cancer, and to elect treatments for instance based on likely response. These identified HRPC-related molecules also can serve as therapeutic targets, and can be used in methods for identifying, developing and testing therapeutic compounds, including for instance rapamycin derivatives, analogs, and mimetics. It will be apparent that the precise details of the methods described may be varied or modified without departing from the spirit of the described invention. We claim all such modifications and variations that fall within the scope and spirit of the claims below.

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Table 1

GenBank ID	Image Clone ID	Clone Name	Cluster location	Max. Correlation	template max	Ratio change	Direction	Unigene	Hs Title	Locus ID	Gene Symbol
AA486027	843249	Human transcriptional repressor (NAB1) NAB1 mRNA, complete cds	1.01	0.85	1	3.18	I	Hs.107474	NGFI-A binding protein 1 (ERG1 binding protein 1)	4664	NAB1
AA463204	796904	Human LOT1 mRNA, complete cds	1.02	0.87	1	2.38	I	Hs.75825	pleomorphic adenoma gene-like 1	5325	PLAGL1
AA733203	399604	BETA-HEXOSAMINIDASE ALPHA CHAIN PRECURSOR	1.04	0.71	1	2.01	I	Hs.119403	hexosaminidase A (alpha polypeptide)	3073	HEXA
AA453926	788205	SRY (sex determining region Y)-box 4	1.04	0.82	1	2.27	I	Hs.83484	SRY (sex determining region Y)-box 4	6659	SOX4
H96235	260303	V-ets avian erythroblastosis virus E26 oncogene homolog 2	1.07	0.78	1	2.61	I	Hs.85146	v-ets avian erythroblastosis virus E26 oncogene homolog 2	2114	ETS2
N62244	287745	Human TAR RNA loop binding protein (TRP-185) mRNA, complete cds	1.07	0.73	1	2.66	I	Hs.151518	TAR (HIV) RNA-binding protein 1	6894	TARBP1
AA460265	796542	ETS-RELATED PROTEIN ERM	1.08	0.82	1	2.08	I	Hs.43697	ets variant gene 5 (ets-related molecule)	2119	ETV5
N49899 N50002	243653	ESTs	1.09	0.75	1	3.31	I	Hs.46981 Hs.1813	ESTs solute carrier family 18 (vesicular monoamine), member 2		SLC18A2
AA043228	486787	Calponin 3, acidic	1.15	0.71	1	3.35	I	Hs.5318	ESTs		
R27712 R27711	134495	ESTs	1.16	0.75	1	2.04	I	Hs.205929 Hs.206066	ESTs ESTs		
AA460679	796680	Human mRNA for CMP-sialic acid transporter, complete cds	1.17	0.83	1	2.00	I	Hs.82921	Homo sapiens mRNA for CMP-sialic acid transporter, complete cds		
AA778198	448386	PRE-B-CELL LEUKEMIA TRANSCRIPTION FACTOR-3	1.17	0.92	1	2.33	I	Hs.171680	pre-B-cell leukemia transcription factor 3	5090	PBX3
N74131	298417	INTESTINAL TREFOIL FACTOR PRECURSOR	1.24	0.95	1	6.13	I	Hs.82961	Human secretory protein (P1.B) mRNA, complete cds		
R02295 R02294	124578	ESTs	1.24	0.90	1	2.28	I	Hs.91226 Hs.204092	ESTs ESTs		

Table 1

GenBank ID	Image Clone ID	Clone Name	Cluster location	Max. Correlation	template max	Ratio change	Direction	Unigene	Hs Title	Locus ID	Gene Symbol
R73608	141562	Human putative monocarboxylate transporter (MCT) mRNA, complete cds	1.27	0.98	1	2.41	I	Hs.23590	solute carrier family 16 (monocarboxylic acid transporters), member 4	9122	SLC16A4
AA283090	713145	CD44 antigen (cell adhesion molecule)	1.29	0.72	1	3.45	I	Hs.169610	CD44 antigen (homing function and Indian blood group system)	960	CD44
AA481543 AA481608	815284	Peptidase D	1.31	0.95	1	3.11	I	Hs.73947 Hs.73947	peptidase D peptidase D		PEPD PEPD
H19440	51408	ZAKI-4 mRNA in human skin fibroblast, complete cds	1.31	0.91	1	2.96	I	Hs.156007	thyroid hormone-responsive (skin fibroblasts)	10231	ZAKI-4
T66053	22012	Human ETS2 gene	1.32	0.75	1	2.51	I	Hs.119177	ADP-ribosylation factor 3	377	ARF3
W73144	344589	Lymphocyte cytosolic protein 1 (L-plastin)	1.32	0.75	1	2.66	I	Hs.76506	ESTs		
AA487623	839101	Cardiac gap junction protein	1.34	0.95	1	4.53	I	Hs.74471	gap junction protein, alpha 1, 43kD (connexin 43)	2697	GJA1
AA088258	511091	Human protein immuno-reactive with anti-PTH polyclonal antibodies mRNA, partial cds	1.35	0.90	1	2.39	I	Hs.44566	Human protein immuno-reactive with anti-PTH polyclonal antibodies mRNA, partial cds		
AA009609	365515	Fibroblast growth factor 7 (keratinocyte growth factor)	1.35	0.82	1	2.43	I	Hs.164568	fibroblast growth factor 7 (keratinocyte growth factor)	2252	FGF7
R77126	144042	ESTs	1.36	0.81	1	2.10	I	Hs.11668	ESTs		
R47979	153411	HLA CLASS II HISTOCOMPATIBILITY ANTIGEN, DR ALPHA CHAIN PRECURSOR	1.38	0.89	1	12.08	I	Hs.76807	Human HLA-DR alpha-chain mRNA		
AA668470	853809	Homo sapiens mRNA for RGS5, complete cds	1.40	0.78	1	2.65	I	Hs.24950	regulator of G-protein signalling 5	8490	RGS5
AA629603	884783	Human PTPL1-associated RhoGAP mRNA, complete cds	1.40	0.78	1	3.05	I	Hs.70983	PTPL1-associated RhoGAP 1	9411	PARG1

Table 1

GenBank ID	Image Clone ID	Clone Name	Cluster location	Max. Correlation template max		Ratio change	Direction	Unigene	Hs Title	Locus ID	Gene Symbol
T59334	75254	Homo sapiens cysteine and glycine-rich protein 2 (CSRP2) mRNA, complete cds	1.42	0.80	1	2.43	I	Hs.10526	cysteine and glycine-rich protein 2 (LIM domain only, smooth muscle)	1466	CSRP2
T80923	109314	ESTs	1.42	0.72	1	2.18	I	Hs.143654	ESTs		
N63943	293925	Lysozyme	1.43	0.81	1	2.43	I	Hs.177746	lysozyme (renal amyloidosis)	4069	LYZ
AA599177	949938	Cystatin C (amyloid angiopathy and cerebral hemorrhage)	1.44	0.81	1	2.20	I	Hs.135084	cystatin C (amyloid angiopathy and cerebral hemorrhage)	1471	CST3
R27432	132835	ESTs	1.45	0.77	1	3.29	I	Hs.114055	ESTs		
AA428893	773567	Protein tyrosine phosphatase, non-receptor type 2	1.93	0.78	2	2.52	I	Hs.82829	protein tyrosine phosphatase, non-receptor type 2	5771	PTPN2
W80632	415529	Human BRCA2 region, mRNA sequence CG006	1.94	0.80	2	2.28	I	Hs.110630	Human BRCA2 region, mRNA sequence CG006		
AA464417	809910	INTERFERON-INDUCIBLE PROTEIN 1-8U	1.95	0.76	2	2.18	I	Hs.182241	Human 1-8U gene from interferon-inducible gene family	10410	
R70541	142326	ESTs	1.96	0.72	2	2.48	I	Hs.23282	ESTs		
R63900	139660	ESTs	1.98	0.82	2	2.21	I	Hs.28456	ESTs		
AA701554	435855	H.sapiens mRNA for hcgVIII protein	1.99	0.93	2	2.48	I	Hs.153618	H.sapiens mRNA for hcgVIII protein		
N39161	243816	CD36 antigen (collagen type I receptor, thrombospondin receptor)	2.00	0.88	2	4.46	I	Hs.75613	CD36 antigen (collagen type I receptor, thrombospondin receptor)	948	CD36
AA102107	489839	Glutamyl aminopeptidase (aminopeptidase A)	2.03	0.82	2	5.15	I	Hs.291	glutamyl aminopeptidase (aminopeptidase A)	2028	ENPEP
R22335	130895	ESTs	2.04	0.85	2	2.08	I	Hs.90790	ESTs		
N30868 N56693	258120	Cytochrome c oxidase subunit VIIb	2.04	0.79	2	4.67	I	Hs:43936 Hs.75752	ESTs cytochrome c oxidase subunit VIIb		COX7B
R53935 R53330	39920	P glycoprotein 3 multiple drug resistance 3	2.05	0.88	2	3.05	I	Hs.172769 Hs.73812	ESTs P glycoprotein 3 multiple drug resistance 3		PGY3
W69954	343867	Allograft inflammatory factor 1	2.06	0.83	2	3.49	I	Hs.76364	allograft inflammatory factor 1	199	AIF1
R69277	142139	ESTs	2.07	0.95	2	2.47	I	Hs.7905	ESTs		

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Table 1

GenBank ID	Image Clone ID	Clone Name	Cluster location	Max. Correlation	template max	Ratio change	Direction	Unigene	Hs Title	Locus ID	Gene Symbol
AA872383	1472735	Human metallothionein-Ie gene (hMT-Ie)	2.07	0.79	2	2.42	I	Hs.74170	metallothionein 1E (functional)	4493	MT1E
AA148641	503083	Homo sapiens homeobox protein MEIS2 (MEIS2) mRNA, partial cds	2.08	0.94	2	3.99	I	Hs.104105	Meis (mouse) homolog 2	4212	MEIS2
AA460986 AA461293	796147	Human GPI-H mRNA, complete cds	2.09	0.71	2	2.06	I	Hs.177 Hs.205655	phosphatidylinositol glycan, class H ESTs	5283	PIGH
AA456321	813179	Insulin-like growth factor 1 (somatomedia C)	2.09	0.89	2	2.77	I	Hs.85112	insulin-like growth factor 1 (somatomedin C)	3479	IGF1
AA401972	741891	Human RalGDS-like 2 (RGL2) mRNA, partial cds	2.09	0.75	2	2.24	I	Hs.170160	GDS-related protein	9264	HKE1.5
AA682293	461727	Homo sapiens phenylalanine hydroxylase (PAH) mutant Q20stop mRNA	2.11	0.98	2	2.77	I	Hs.1870	phenylalanine hydroxylase	5053	PAH
AA428778	756968	Human placenta LERK-2 (EPLG2) mRNA, complete cds	2.11	0.94	2	2.05	I	Hs.144700	ephrin-B1	1947	EFNB1
AA481026	814636	H.sapiens hbrm mRNA	2.12	0.71	2	2.17	I	Hs.199211	ESTs		
AA496809	897667	SNF2 (sucrose nonfermenting, yeast, homolog)-like 1	2.13	0.97	2	3.73	I	Hs.152292	SWI/SNF related, matrix associated, actin dependent regulator of chromatin, subfamily a, member 1	6594	SMARCA1
N80129	297392	Metallothionein 1L	2.13	0.86	2	2.80	I	Hs.94360	metallothionein 1L	4500	MT1L
W39618	322723	ESTs	2.13	0.85	2	2.28	I	Hs.93231	ESTs		
T66799	66322	CD3G antigen, gamma polypeptide (TiT3 complex)	2.13	0.93	2	5.33	I	Hs.2259	CD3G antigen, gamma polypeptide (TiT3 complex)	917	CD3G
N72715	295729	Homo sapiens mRNA for translational inhibitor protein p14.5	2.14	0.79	2	2.55	I	Hs.18426	translational inhibitor protein p14.5	10247	UK114
N25338 N31452	265645	Human mRNA for histamine N-methyltransferase, complete cds	2.15	0.70	2	2.05	I	Hs.161090 Hs.81182	EST Human histamine N-methyltransferase (HNMT) mRNA, complete cds		

Table 1

GenBank ID	Image Clone ID	Clone Name	Cluster location	Max. Correlation	template max	Ratio change	Direction	Unigene	Hs Title	Locus ID	Gene Symbol
R76498	143756	ESTs	2.16	0.96	2	2.43	I	Hs.93097	ESTs		
R89567 R88884	195340	Group-specific component (vitamin D binding protein)	2.18	0.93	2	3.77	I	Hs.174319 Hs.34212	ESTs Vitamin D binding protein {Alu sequence, exon 9, intron 8} [human, blood cells, Genomic, 469 nt]		
T50788 T50951	78294	UDP glucuronosyltransferase precursor (UGT2B15)	2.19	0.98	2	6.32	I	Hs.203276 Hs.150207	UDP glycosyltransferase 2 family, polypeptide B15 UDP glycosyltransferase 2 family, polypeptide B15		UGT2B15 UGT2B15
N74236	296880	Membrane protein, palmitoylated 1 (55kD)	2.19	0.97	2	2.92	I	Hs.1861	membrane protein, palmitoylated 1 (55kD)	4354	MPP1
AA290738	713922	Glutathione S-transferase M4	2.20	0.95	2	2.85	I	Hs.154159	glutathione S-transferase M1	2944	GSTM1
H56088	203772	ESTs	2.21	0.96	2	2.87	I	Hs.34498	ESTs		
AA074224	383188	Recoverin	2.22	0.70	2	2.11	I	Hs.80539	recoverin	5957	RCV1
AA682631	431296	Protein phosphatase 3 (formerly 2B), catalytic subunit, alpha isoform (calcineurin A alpha){alternative products}	2.22	0.97	2	2.57	I	Hs.92	protein phosphatase 3 (formerly 2B), catalytic subunit, alpha isoform (calcineurin A alpha)	5530	PPP3CA
AA489331	842939	Human dsRNA adenosine deaminase DRADA2b (DRADA2b) mRNA, complete cds	2.23	0.74	2	2.11	I	Hs.85302	adenosine deaminase, RNA-specific, B1 (homolog of rat RED1)	104	ADARB1
W81191	347434	Homo sapiens nucleolar autoantigen No55 mRNA, complete cds	2.24	0.82	2	2.59	I	Hs.121927	Homo sapiens nucleolar autoantigen No55 mRNA, complete cds		
N92319	308041	Glycoprotein Ib (platelet), beta polypeptide	2.25	0.76	2	2.77	I	Hs.3847	peanut (Drosophila)-like 1	5413	PNUTL1
H53340	202535	Human (clone 14VS) metallothionein-IG (MT1G) gene, complete cds	2.25	0.93	2	2.01	I	Hs.173451	metallothionein 1G	4495	MT1G

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Table 1

GenBank ID	Image Clone ID	Clone Name	Cluster location	Max. Correlation	template max	Ratio change	Direction	Unigene	Hs Title	Locus ID	Gene Symbol
T67092 T67093	66582	ESTs	2.27	0.88	2	2.68	I	Hs.13035 Hs.154424	ESTs deiodinase iodothyronine, type II		DIO2
AA460282	795730	Homo sapiens mRNA for Efs1, complete cds	2.28	0.70	2	2.37	I	Hs.24587	signal transduction protein (SH3 containing)	10278	EFS2
N63646	292770	ESTs	2.30	0.80	2	2.10	I	Hs.172084	ESTs		
AA453175	788107	Homo sapiens amphiphysin II mRNA, complete cds	2.31	0.78	2	3.49	I	Hs.206051	Homo sapiens box-dependent myc-interacting protein isoform BIN1-10-13 (BIN1) mRNA, complete cds		
R12373 R09781	128143	Paraoxonase 1	2.34	0.77	2	6.22	I	Hs.196620 Hs.194738	ESTs H.sapiens serum paraoxonase (PON) mRNA, 3' end		
H77597	214162	H.sapiens mRNA for metallothionein	2.35	0.78	2	2.70	I	Hs.2667	metallothionein 1H	4496	MT1H
N70841	298231	Homo sapiens mRNA for GABA-BR1a (hGB1a) receptor	2.35	0.80	2	2.91	I	Hs.167017	gamma-aminobutyric acid (GABA) B receptor, 1 "confirmed"	2550	GABBR1
AA700222	453183	Homo sapiens SH3-containing adaptor molecule-1 mRNA, complete cds	2.37	0.82	2	2.38	I	Hs.33787	vinexin beta (SH3-containing adaptor molecule-1)	10174	SCAM-1
AA156802	502518	Laminin, beta 2 (laminin S)	2.37	0.82	2	2.70	I	Hs.90291	laminin, beta 2 (laminin S)	3913	LAMB2
AA482637	755054	Human putative transmembrane receptor IL-1Rrp mRNA, complete cds	2.41	0.76	2	3.27	I	Hs.159301	interleukin 18 receptor 1	8809	IL18R1
W85710	415978	Human carnitine palmitoyltransferase I (CPTI) mRNA, complete cds	2.42	0.79	2	2.17	I	Hs.29331	carnitine palmitoyltransferase I, liver	1374	CPT1A
AA219061	630013	DNA repair protein MSH2	2.90	0.83	3	4.63	I	Hs.78934	mutS (E. coli) homolog 2 (colon cancer, nonpolyposis type 1)	4436	MSH2
AA453742	813678	EXCITATORY AMINO ACID TRANSPORTER 1	2.90	0.87	3	2.53	I	Hs.75379	solute carrier family 1 (glial high affinity glutamate transporter),	6507	SLC1A3

Table 1

GenBank ID	Image Clone ID	Clone Name	Cluster location	Max. Correlation	template max	Ratio change	Direction	Unigene	Hs Title	Locus ID	Gene Symbol
H61243	236034	Uncoupling protein 2 (mitochondrial, proton carrier)	2.90	0.85	3	2.23	I	Hs.80658	uncoupling protein 2 (mitochondrial, proton carrier)	7351	UCP2
N30553	257162	Pregnancy-specific beta-1 glycoprotein 4	2.91	0.89	3	3.30	I	Hs.206128	pregnancy specific beta-1-glycoprotein 4	5672	PSG4
AA447797	813841	Plasminogen activator, tissue type (t-PA)	2.92	0.84	3	3.10	I	Hs.173736	plasminogen activator, tissue	5327	PLAT
AA430552	770074	Homo sapiens proline-rich Gla protein 2 (PRGP2) mRNA, complete cds	2.92	0.71	3	2.20	I	Hs.35101	proline-rich Gla (G-carboxglutamic acid) polypeptide 2	5639	PRRG2
N21576	266146	Human mitochondrial 1,25-dihydroxyvitamin D3 24-hydroxylase mRNA, complete cds	2.92	0.89	3	3.88	I	Hs.198092	ESTs		
AA056042	377692	Human microfibril-associated glycoprotein-2 MAGP-2 mRNA, complete cds	2.92	0.98	3	2.54	I	Hs.58882	Microfibril-associated glycoprotein-2	8076	MAGP2
AA496565	755952	Homo sapiens microsomal glutathione S-transferase 3 (MGST3) mRNA, complete cds	2.93	0.97	3	2.11	I	Hs.200480	plexin 5	5364	PLXN5
W03787	297063	ESTs	2.93	0.89	3	2.18	I	Hs.90638	ESTs		
AA486321	840511	Vimentin	2.94	0.84	3	3.02	I	Hs.2064	vimentin	7431	VIM
W93369	415084	Cholinergic receptor, nicotinic, alpha polypeptide 7	2.95	0.96	3	4.09	I	Hs.167418	ESTs		
AA487590	841695	Human BRCA2 region, mRNA sequence CG018	2.95	0.93	3	5.06	I	Hs.22174	Human BRCA2 region, mRNA sequence CG018		
H45455	183462	Homo sapiens alpha-mannosidase (6A8) mRNA, complete cds	2.95	0.76	3	2.05	I	Hs.26232	Homo sapiens alpha mannosidase 6A8B (6a8b) mRNA, complete cds		

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GenBank ID	Image Clone ID	Clone Name	Cluster location	Max. Correlation	template max	Ratio change	Direction	Unigene	Hs Title	Locus ID	Gene Symbol
AA421284	731051	Homo sapiens clone 23619 phosphoprotein mRNA, partial cds	2.96	0.91	3	2.71	I	Hs.196202	Homo sapiens clone 23619 phosphoprotein mRNA, partial cds		
T64223	80221	Carboxypeptidase A3 (mast cell)	2.96	0.92	3	2.53	I	Hs.646	carboxypeptidase A3 (mast cell)	1359	CPA3
AA437291	758222	Hydroxysteroid (17-beta) dehydrogenase 3	2.97	0.80	3	6.52	I	Hs.477	hydroxysteroid (17-beta) dehydrogenase 3	3293	HSD17B3
T50282	71116	TISSUE FACTOR PATHWAY INHIBITOR PRECURSOR	2.97	0.73	3	2.41	I	Hs.170279	tissue factor pathway inhibitor (lipoprotein-associated coagulation inhibitor)	7035	TFPI
R36006	137139	ESTs	2.97	0.81	3	5.53	I	Hs.64065	ESTs		
R34604 R34603	136557	ESTs	2.97	0.87	3	3.30	I	Hs.24822 Hs.198609	ESTs ESTs		
H60824 H60910	205239	PROTEIN KINASE C, THETA TYPE	2.98	0.74	3	7.36	I	Hs.195553 Hs.196882	ESTs ESTs		
AA045699	509731	TUMOR-ASSOCIATED ANTIGEN CO-029	2.98	0.98	3	3.17	I	Hs.84072	transmembrane 4 superfamily member 3	7103	TM4SF3
H63161	208718	Annexin I (lipocortin I)	2.98	0.92	3	2.90	I	Hs.78225	annexin I (lipocortin I)	301	ANX1
AA485865	840460	Interleukin 7 receptor	2.98	0.84	3	4.56	I	Hs.109703	interleukin 7 receptor	3575	IL7R
T91100	111750	ESTs	2.98	0.97	3	4.55	I	Hs.15702	ESTs		
AA282134	711918	H.sapiens mRNA for glutamine cyclotransferase	2.98	0.90	3	3.13	I	Hs.79033	H.sapiens mRNA for glutamine cyclotransferase		
AA454854	809998	ALPHA-AMYLASE 2B PRECURSOR	2.98	0.87	3	4.88	I	Hs.75733	ESTs		
AA447079	784296	Mineralocorticoid receptor (aldosterone receptor)	2.98	0.98	3	3.31	I	Hs.1790	mineralocorticoid receptor (aldosterone receptor)	4306	MLR
AA455925	813266	Homo sapiens skeletal muscle LIM-protein FHL1 mRNA, complete cds	2.99	0.82	3	2.26	I	Hs.75329	four and a half LIM domains 1	2273	FHL1

Table 1

GenBank ID	Image Clone ID	Clone Name	Cluster location	Max. Correlation	template max	Ratio change	Direction	Unigene	Hs Title	Locus ID	Gene Symbol
N27179	257422	Human mRNA for BST-1, complete cds	2.99	0.79	3	3.46	I	Hs.169998	bone marrow stromal cell antigen 1	683	BST1
T73556	82734	Long chain fatty acid acyl-coA ligase	2.99	0.87	3	3.27	I	Hs.154890	fatty-acid-Coenzyme A ligase, long-chain 1	2179	FACL1
AA486482	811161	Homo sapiens TNF-alpha stimulated ABC protein (ABC50) mRNA, complete cds	2.99	0.85	3	2.02	I	Hs.9573	ATP-binding cassette 50 (TNF-alpha stimulated)	23	ABC50
W37864	322160	Phosphatase and tensin homolog (mutated in multiple advanced cancers 1)	2.99	0.91	3	3.88	I	Hs.10712	phosphatase and tensin homolog (mutated in multiple advanced cancers 1)	5728	PTEN
AA670134	844725	Human mRNA for TPRD, complete cds	2.99	0.91	3	2.65	I	Hs.118174	tetratricopeptide repeat domain 3	7267	TTC3
R94153	276091	Inositol 1,4,5-trisphosphate 3-kinase B	2.99	0.83	3	2.51	I	Hs.78877	inositol 1,4,5-trisphosphate 3-kinase B	3707	ITPKB
H45976	177772	Human cadherin-associated protein-related (cap-r) mRNA, complete cds	2.99	0.86	3	3.34	I	Hs.150917	Human cadherin-associated protein-related (cap-r) mRNA, complete cds		
AA682684	461804	Protein tyrosine phosphatase, non-receptor type 3	3.00	0.75	3	5.06	I	Hs.153932	protein tyrosine phosphatase, non-receptor type 3	5774	PTPN3
AA598668	898258	N-CHIMAERIN	3.00	0.97	3	2.05	I	Hs.169965	chimerin (chimaerin) 1	1123	CHN1
N58558 N78159	248412	Homo sapiens kallistatin (PI4) mRNA, complete cds	3.00	0.78	3	3.37	I	Hs.159628 Hs.179779	protease inhibitor 4 (kallistatin) ribosomal protein L37	5267	PI4 RPL37
W69790	344139	Human ovarian cancer downregulated myosin heavy chain homolog (Doc1) mRNA, complete cds	3.00	0.73	3	3.43	I	Hs.15432	Human ovarian cancer downregulated myosin heavy chain homolog (Doc1) mRNA, complete cds		
AA460688	796689	Homo sapiens FMRFamide-related prepropeptide mRNA, complete cds	3.00	0.95	3	2.41	I	Hs.104555	FMFRamide-related peptide precursor	8620	FMRFAL
H86554	223350	Ceruloplasmin (ferroxidase)	3.01	0.95	3	8.30	I	Hs.204819	ceruloplasmin (ferroxidase)	1356	CP

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GenBank ID	Image Clone ID	Clone Name	Cluster location	Max. Correlation	template max	Ratio change	Direction	Unigene	Hs Title	Locus ID	Gene Symbol
T56221	78353	Human metallothionein (MT)I-F gene	3.01	0.99	3	2.56	I	Hs.110440	Human metallothionein (MT)I-F gene		
AA128328	502832	Retinoblastoma-binding protein 1{alternative products}	3.01	0.91	3	4.08	I	Hs.91797	retinoblastoma-binding protein 1	5926	RBBP1
H95959	250654	SPARC/osteonectin	3.01	0.94	3	2.92	I	Hs.111779	secreted protein, acidic, cysteine-rich (osteonectin)	6678	SPARC
H72028	214990	GELSOLIN PRECURSOR, PLASMA	3.01	0.93	3	2.57	I	Hs.80562	gelsolin (amyloidosis, Finnish type)	2934	GSN
AA447692	813611	Human homeobox gene (clone HHO.c13)	3.01	0.80	3	8.69	I	Hs.197863	ESTs		
R66310	140806	Peptidylglycine alpha-amidating monooxygenase	3.01	0.95	3	3.54	I	Hs.83920	peptidylglycine alpha-amidating monooxygenase	5066	PAM
AA865729	1469234	H.sapiens mRNA (ocular albinism type 1 related)	3.01	0.94	3	2.20	I	Hs.74124	ocular albinism 1 (Nettleship-Falls)	4935	OA1
AA448400	781362	Human plectin (PLEC1) mRNA, complete cds	3.01	0.93	3	2.64	I	Hs.194860	ESTs		
AA453759	813698	Homo sapiens Sprouty 2 (SPRY2) mRNA, complete cds	3.01	0.99	3	2.40	I	Hs.18676	sprouty (Drosophila) homolog 2	10253	SPRY2
AA056232	377731	Glutathione S-transferase M5	3.01	0.95	3	2.30	I	Hs.75652	glutathione S-transferase M5	2949	GSTM5
T64057	79782	Human mRNA for DB1, complete cds	3.02	0.88	3	2.15	I	Hs.6557	ESTs		
AA430345	768644	ZONA PELLUCIDA SPERM-BINDING PROTEIN 3A PRECURSOR	3.02	0.81	3	2.13	I	Hs.144645	Human POM-ZP3 mRNA, complete cds		
AA405569	772425	Human fibroblast activation protein mRNA, complete cds	3.02	0.91	3	3.00	I	Hs.418	fibroblast activation protein, alpha	2191	FAP
H02340	150702	Homeo box B5 (2.1 protein)	3.02	0.88	3	2.07	I	Hs.22554	homeo box B5	3215	HOXB5

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GenBank ID	Image Clone ID	Clone Name	Cluster location	Max. Correlation	template max	Ratio change	Direction	Unigene	Hs Title	Locus ID	Gene Symbol
N53031	246430	UDP-GLUCURONOSYLTRANSFERASE 2B4 PRECURSOR, MICROSOMAL	3.03	0.99	3	3.91	I	Hs.89691	Homo sapiens UDP glucuronosyltransferase 2B4 precursor (UGT2B4) mRNA, UGT2B4*L109, L396 variant allele, complete cds		
H64888	210494	ESTs	3.04	0.97	3	2.34	I	Hs.38327	ESTs		
T95657	120681	ESTs	3.04	0.83	3	2.26	I	Hs.194292	ESTs		
R26163	132140	ESTs	3.04	0.81	3	4.53	I	Hs.93961	ESTs		
AA676223	431501	H.sapiens mRNA for pristanoyl-CoA oxidase	3.04	0.80	3	2.29	I	Hs.12773	acyl-Coenzyme A oxidase 3, pristanoyl	8310	ACOX3
AA026470	366966	ESTs	3.04	0.92	3	2.26	I	Hs.27865	ESTs (3' of PTEN)		
R45413		Human transmembrane 4 superfamily protein (SAS)						Hs.50984	sarcoma amplified sequence		
R25074	35105	mRNA, complete cds	3.05	0.83	3	2.38	I	Hs.146261	ESTs	6302	SAS
R99084	201393	ESTs	3.05	0.76	3	2.41	I	Hs.36069	ESTs		
R35979	137096	ESTs	3.05	0.93	3	3.29	I	Hs.25056	EST		
AA779165	453005	Human ADP-ribosylation factor-like protein 4 mRNA, complete cds	3.05	0.94	3	2.80	I	Hs.177743	ESTs		
R23227	131050	ESTs	3.05	0.97	3	2.71	I	Hs.23479	ESTs		
AA496916		Dual-specificity tyrosine-(Y)-phosphorylation regulated						Hs.66185	Homo Sapiens mRNA, partial cDNA sequence from cDNA selection, DCR1-16.0 dual-specificity tyrosine-(Y)-phosphorylation regulated kinase		
AA497079	897485	kinase	3.05	0.94	3	2.83	I	Hs.75842			DYRK1
AA446316		Proopiomelanocortin (adrenocorticotropin/ beta-lipotropin/ alpha-melanocyte stimulating hormone/ beta-melanocyte stimulating						Hs.191622	ESTs proopiomelanocortin (adrenocorticotropin/ beta-lipotropin/ alpha-melanocyte stimulating hormone/ beta-melanocyte		
AA446498	781233	hormone/ beta-endorphin)	3.05	0.72	3	3.70	I	Hs.1897	stimulating hormone/ beta-endorphin)		POMC

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GenBank ID	Image Clone ID	Clone Name	Cluster location	Max. Correlation	template max	Ratio change	Direction	Unigene	Hs Title	Locus ID	Gene Symbol
AA424938	768246	Glucose-6-phosphate dehydrogenase	3.06	0.99	3	4.17	I	Hs.80206	glucose-6-phosphate dehydrogenase	2539	G6PD
AA598652	897906	Sialyltransferase 1 (beta-galactoside alpha-2,6-sialytransferase)	3.06	0.92	3	3.78	I	Hs.2554	sialyltransferase 1 (beta-galactoside alpha-2,6-sialytransferase)	6480	SIAT1
N53169	246765	Apolipoprotein C-III	3.06	0.96	3	2.71	I	Hs.73849	apolipoprotein C-III	345	APOC3
AA845156	1412481	Serine protease inhibitor, Kazal type 1	3.06	0.95	3	3.77	I	Hs.181286	serine protease inhibitor, Kazal type 1	6690	SPINK1
T85091	111150	ESTs	3.06	0.86	3	2.56	I	Hs.205999	ESTs		
R02586	124071	ESTs	3.06	0.97	3	3.41	I	Hs.19404	ESTs		
R60343	42070	5' nucleotidase (CD73)	3.06	0.97	3	3.37	I	Hs.153952	5' nucleotidase (CD73)	4907	NT5
AA862371	1455976	INTERFERON-INDUCIBLE PROTEIN 1-8D	3.06	0.94	3	2.46	I	Hs.174195	Human 1-8D gene from interferon-inducible gene family		
T67549	66982	Plasminogen-like protein	3.07	0.94	3	2.54	I	Hs.75576	plasminogen	5340	PLG
AA018780	362483	Spectrin, beta, non-erythrocytic 1	3.08	0.90	3	4.63	I	Hs.107164	spectrin, beta, non-erythrocytic 1	6711	SPTBN1
AA062993	382195	Myosin VIIA (Usher syndrome 1B (autosomal recessive, severe))	3.09	0.82	3	3.43	I	Hs.95361	myosin VIIA (Usher syndrome 1B (autosomal recessive, severe))	4647	MYO7A
R93124	196992	Dihydrodiol dehydrogenase	3.10	0.81	3	2.66	I	Hs.201967	aldo-keto reductase family 1, member C1 (dihydrodiol dehydrogenase 1; 20-alpha (3-alpha)-hydroxysteroid dehydrogenase)	1645	AKR1C1
AA115877	564621	H.sapiens mRNA for protease inhibitor 12 (PI12; neuroserpin)	3.10	0.86	3	2.27	I	Hs.78589	protease inhibitor 12 (neuroserpin)	5274	PI12
AA458922	814409	Human lysophosphatidic acid acyltransferase-alpha mRNA, complete cds	3.54	0.70	4	2.04	I	Hs.93728	pre-B-cell leukemia transcription factor 2	5089	PBX2
AA148737	504763	Syndecan 4 (amphiglycan, ryudocan)	3.55	0.71	4	2.92	I	Hs.72082	syndecan 4 (amphiglycan, ryudocan)	6385	SDC4

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GenBank ID	Image Clone ID	Clone Name	Cluster location	Max. Correlation	template max	Ratio change	Direction	Unigene	Hs Title	Locus ID	Gene Symbol
R75820	143519	FK506-BINDING PROTEIN PRECURSOR	3.60	0.74	4	2.39	I	Hs.23476	FK506-binding protein 2 (13kD)	2286	FKBP2
T69304	82903	Homo sapiens tapasin (NGS-17) mRNA, complete cds	3.60	0.80	4	2.73	I	Hs.179600	TAP binding protein (tapasin)	6892	TAPBP
H77652	234736	Human transcription factor hGATA-6 mRNA, complete cds	3.62	0.81	4	2.15	I	Hs.50924	GATA-binding protein 6	2627	GATA6
AA455043	812246	Holocarboxylase synthetase (biotin-[proprionyl-Coenzyme A-carboxylase (ATP-hydrolysing)] ligase)	3.63	0.80	4	2.56	I	Hs.79375	holocarboxylase synthetase (biotin-[proprionyl-Coenzyme A-carboxylase (ATP-hydrolysing)] ligase)	3141	HLCS
R07594	125685	ESTs	3.66	0.78	4	2.17	I	Hs.172620	ESTs		
AA703250	436155	Homo sapiens U4/U6 small nuclear ribonucleoprotein hPrp4 mRNA, complete cds	3.66	0.78	4	2.01	I	Hs.8551	PRP4/STK/WD splicing factor	9128	HPRP4P
AA598610	898219	Mesoderm specific transcript (mouse) homolog	3.67	0.73	4	2.01	I	Hs.79284	mesoderm specific transcript (mouse) homolog	4232	MEST
AA431885	773637	Human mRNA for MNK1, complete cds	3.67	0.84	4	2.60	I	Hs.5591	MAP kinase-interacting serine/threonine kinase 1	8569	MKNK1
N24880 N36299	269878	Cytochrome c oxidase subunit X (heme A: farnesyltransferase	3.70	0.74	4	2.61	I	Hs.138660 Hs.77513	ESTs cytochrome c oxidase subunit X (heme A: farnesyltransferase)		COX10
AA001614	427812	Insulin receptor	3.71	0.87	4	2.53	I	Hs.89695	insulin receptor	3643	INSR
AA634103	868368	Human thymosin beta-4 mRNA, complete cds	3.71	0.76	4	2.38	I	Hs.75968	thymosin, beta 4, X chromosome	7114	TMSB4X
	81475	Homo sapiens Notch3 (NOTCH3) mRNA, complete cds	3.71	0.79	4	2.24	I				
AA447632	784910	Human fetus brain mRNA for membrane glycoprotein M6, complete cds	3.72	0.91	4	2.22	I	Hs.75819	glycoprotein M6A	2823	GPM6A

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AA668595	859359	Homo sapiens Pig3 (PIG3) mRNA, complete cds	3.73	0.85	4	2.20	I	Hs.50649	quinone oxidoreductase homolog	9540	PIG3
AA456570	809353	H.sapiens mRNA for interferon regulatory factor 3	3.74	0.76	4	2.33	I	Hs.75254	interferon regulatory factor 3	3661	IRF3
AA436564	753069	Human cellular proto-oncogene (c-mer) mRNA, complete cds	3.74	0.77	4	2.35	I	Hs.78941	c-mer proto-oncogene tyrosine kinase	10461	MERTK
AA055504 AA055503	377320	Human zinc-finger protein mRNA, complete cds	3.75	0.80	4	2.13	I	Hs.173067 Hs.204377	ESTs Human zinc-finger protein mRNA, complete cds		
AA872420	1472775	Human COL8A1 mRNA for alpha 1(VIII) collagen	3.75	0.72	4	2.88	I	Hs.114599	collagen, type VIII, alpha 1	1295	COL8A1
R22189	130826	ESTs	3.75	0.95	4	3.29	I	Hs.23349	ESTs NO HOMOLOGY TO		
H09111	46284	Human infant brain mRNA, clone 13cDNA73	3.76	0.94	4	2.04	I	Hs.181304	putative gene product	10129	13CDNA73
AA130579	586685	Human galectin-4 (GAL4) mRNA, complete cds	3.76	0.92	4	2.23	I	Hs.5302	lectin, galactoside-binding, soluble, 4 (galectin 4)	3960	LGALS4
AA485748	811162	Fibromodulin	3.77	0.89	4	4.10	I	Hs.230	fibromodulin	2331	FMOD
H16824	50680	Human Smg GDS-associated protein SMAP mRNA, complete cds	3.79	0.97	4	2.01	I	Hs.171374	ESTs		
AA486570	840990	Glutathione S-transferase M4	3.80	0.94	4	2.30	I	Hs.82891	glutathione S-transferase M4	2948	GSTM4
AA885311	1461664	Butyrylcholinesterase	3.81	0.98	4	2.22	I	Hs.1327	butyrylcholinesterase	590	BCHE
AA461456	796613	Collagen, type V, alpha	3.81	0.88	4	3.65	I	Hs.82985	collagen, type V, alpha 2	1290	COL5A2
T54166	68977	PROTEASOME COMPONENT MECL-1 PRECURSOR	3.82	0.71	4	2.41	I	Hs.9661	proteasome (prosome, macropain) subunit, beta type, 10	5699	PSMB10
AA461108 AA461424	796198	Eph-related receptor tyrosine kinase ligand 5	3.82	0.97	4	2.08	I	Hs.206183 Hs.30942	ESTs ephrin-B2		EFNB2

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GenBank ID	Image Clone ID	Clone Name	Cluster location	Max. Correlation	template max	Ratio change	Direction	Unigene	Hs Title	Locus ID	Gene Symbol
T65736	80338	Human selenium-binding protein (hSBP) mRNA, complete cds	3.83	0.79	4	2.42	I	Hs.7833	selenium binding protein 1	8991	SELENBP1
AA430668	770394	Human IgG Fc receptor hFcRn mRNA, complete cds	3.84	0.95	4	2.93	I	Hs.160741	Fc fragment of IgG, receptor, transporter, alpha	2217	FCGRT
R62817	138936	ERYTHROCYTE BAND 7 INTEGRAL MEMBRANE PROTEIN	3.84	0.82	4	3.10	I	Hs.160483	erythrocyte membrane protein band 7.2 (stomatin)	2040	EPB72
AA443302								Hs.6838	ESTs		
AA443435	784593	RhoE	3.85	0.88	4	2.64	I	Hs.206166	ESTs		
AA424743								Hs.85155	butyrate response factor 1 (EGF-response factor 1)		
AA495759	768299	H.sapiens ERF-1 mRNA 3' end	3.85	0.86	4	2.43	I	Hs.33905	ESTs	677	BRF1
H72722	232772	Human metallothionein I-B gene	3.86	0.91	4	2.06	I	Hs.36102	ESTs		
R43483	32493	Integrin, alpha 6	3.86	0.93	4	2.05	I	Hs.200466	ESTs		
AA679422	432210	Arginine carboxypeptidase (carboxypeptidase N)	3.87	0.77	4	3.06	I	Hs.2246	carboxypeptidase N, polypeptide 1, 50kD	1369	CPN1
R07296	126858	Sterol O-acyltransferase (acyl-Coenzyme A: cholesterol acyltransferase)	3.87	0.81	4	2.10	I	Hs.14553	sterol O-acyltransferase (acyl-Coenzyme A: cholesterol acyltransferase) 1	6646	SOAT1
N55430	245936	ESTs	3.87	0.86	4	2.65	I	Hs.47962	ESTs		
AA876039	1161775	VILLIN	3.87	0.77	4	2.67	I	Hs.3046	ESTs		
AA486627	840942	Major histocompatibility complex, class II, DP beta 1	3.88	0.93	4	2.07	I	Hs.814	major histocompatibility complex, class II, DP beta 1	3115	HLA-DPB1
AA001449	361974	Pleiotrophin (heparin binding growth factor 8, neurite growth-promoting factor 1)	3.88	0.98	4	2.02	I	Hs.44	pleiotrophin (heparin binding growth factor 8, neurite growth-promoting factor 1)	5764	PTN

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H18950 H19393	51406	H.sapiens mRNA for hepatocyte nuclear factor 4 gamma	3.89	0.87	4	2.73	I	Hs.102867 Hs.202659	ESTs hepatocyte nuclear factor 4, gamma		HNF4G
R52798	41650	Hepatocyte growth factor (hepapoietin A; scatter factor)	3.90	0.89	4	2.97	I	Hs.809	hepatocyte growth factor (hepapoietin A; scatter factor)	3082	HGF
T55870	77728	Human leukemia virus receptor 2 (GLVR2) mRNA, complete cds	3.90	0.73	4	2.23	I	Hs.75867	Human leukemia virus receptor 2 (GLVR2) mRNA, complete cds		
N66177 N99168	278570	Microphthalmia-associated transcription factor	3.90	0.79	4	2.71	I	Hs.82000 Hs.166017	Homo sapiens mRNA for A-type microphthalmia associated transcription factor, complete cds microphthalmia-associated transcription factor		MITF
N35070	271670	Homo sapiens TWEAK mRNA, complete cds	3.91	0.87	4	2.30	I	Hs.26401	tumor necrosis factor (ligand) superfamily, member 12	8742	TNFSF12
R20729	26249	Homo sapiens EEN-B1 mRNA, complete cds	3.92	0.91	4	2.36	I	Hs.75149	SH3-domain GRB2-like 2	6456	SH3GL2
AA464152 AA464217	810331	Homo sapiens quiescin (Q6) mRNA, complete cds	3.92	0.91	4	2.10	I	Hs.77266 Hs.71816	quiescin Q6 v-akt murine thymoma viral oncogene homolog 1	5768	QSCN6 AKT1
AA485665	811088	Human putative EPH-related PTK receptor ligand LERK-8 (Eplg8) mRNA, complete cds	3.92	0.89	4	3.43	I	Hs.26988	ephrin-B3	1949	EFNB3
AA191518	627306	Crystallin beta-B2	3.93	0.88	4	2.08	I	Hs.169286	crystallin, beta B2	1415	CRYBB2
H51574	179890	Arachidonate 5-lipoxygenase	3.93	0.96	4	2.29	I	Hs.89499	arachidonate 5-lipoxygenase	240	ALOX5
T97276	121454	Arachidonate 12-lipoxygenase	3.93	0.92	4	2.34	I	Hs.1200	arachidonate 12-lipoxygenase	239	ALOX12
N63968	289447	POU homeobox protein	3.94	0.83	4	2.22	I	Hs.2815	POU domain, class 6, transcription factor 1	5463	POU6F1

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Table 1

GenBank ID	Image Clone ID	Clone Name	Cluster location	Max. Correlation	template max	Ratio change	Direction	Unigene	Hs Title	Locus ID	Gene Symbol
AA464970	810104	1-PHOSPHATIDYLINOSITOL-4,5-BISPHOSPHATE PHOSPHODIESTERASE BETA 2	3.94	0.70	4	4.25	I	Hs.994	phospholipase C, beta 2	5330	PLCB2
AA446251	774471	Laminin B1 chain	3.94	0.77	4	2.75	I	Hs.82124	laminin, beta 1	3912	LAMB1
AA055350	377252	Adenosine A2b receptor	3.96	0.75	4	2.00	I	Hs.45743	adenosine A2b receptor	136	ADORA2B
AA599173	949928	Human monocytic leukaemia zinc finger protein (MOZ) mRNA, complete cds	3.96	0.81	4	2.23	I	Hs.82210	Human monocytic leukaemia zinc finger protein (MOZ) mRNA, complete cds		
N69540	294881	ESTs	3.96	0.86	4	2.22	I	Hs.17713	ESTs		
N62554	292364	ESTs	3.97	0.94	4	2.12	I	Hs.48565	ESTs		
R71093	142788	Human mRNA for collagen binding protein 2, complete cds	3.97	0.92	4	3.26	I	Hs.9930	collagen-binding protein 2 (colligen 2)	872	CBP2
N53136	246619	Cytochrome P450, subfamily IIC (mephenytoin 4-hydroxylase)	3.97	0.95	4	2.62	I	Hs.174220	cytochrome P450, subfamily IIC (mephenytoin 4-hydroxylase), polypeptide 8	1558	CYP2C8
W77839	345935	Homo sapiens mad protein homolog (hMAD-3) mRNA, complete cds	3.98	0.94	4	5.65	I	Hs.198008	ESTs		
AA630374	854899	H.sapiens mRNA for protein-tyrosine-phosphatase (tissue type: foreskin)	3.98	0.78	4	2.94	I	Hs.180383	dual specificity phosphatase 6	1848	DUSP6
AA427659	770059	Human heparan sulfate proteoglycan (HSPG2) mRNA, complete cds	3.98	0.75	4	2.02	I	Hs.75578	ESTs		
H29256 H29339	49630	Human neuroendocrine/beta-cell-type calcium channel alpha-1 subunit mRNA, complete cds	4.01	0.80	4	2.88	I	Hs.203672 Hs.23838	ESTs calcium channel, voltage-dependent, L type, alpha 1D subunit		CACNA1D
AA425217	773301	Cadherin 3 (P-cadherin)	4.02	0.93	4	4.07	I	Hs.2877	cadherin 3, P-cadherin (placental)	1001	CDH3

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AA429895	781139	Human multidrug resistance-associated protein homolog (MRP3) mRNA, partial cds	4.03	0.95	4	4.84	I	Hs.90786	canicular multispecific organic anion transporter	8714	CMOAT2
AA668821	854338	Chitinase 1	4.03	0.95	4	2.12	I	Hs.154138	chitinase 3-like 2	1117	CHI3L2
AA449738	785967	Homo sapiens protein 4.1-G mRNA, complete cds	4.04	0.92	4	2.04	I	Hs.7857	erythrocyte membrane protein band 4.1-like 2	2037	EPB41L2
AA427468	770388	Homo sapiens hCPE-R mRNA for CPE-receptor, complete cds	4.05	0.81	4	2.05	I	Hs.5372	claudin 4	1364	CLDN4
AA459401	810960	Homo sapiens serine protease-like protease (nes1) mRNA, complete cds	4.05	0.85	4	3.69	I	Hs.198440	ESTs		
AA074222 AA074115	383175	Homo sapiens mRNA for SART-1, complete cds	4.08	0.71	4	2.11	I	Hs.177426 Hs.18813	EST squamous cell carcinoma antigen recognised by T cells		SART1
R10133	128460	ESTs	4.10	0.89	4	2.06	I	Hs.20552	ESTs		
N57553	279970	Adenosine receptor A2	4.12	0.84	4	2.15	I	Hs.1613	adenosine A2a receptor	135	ADORA2A
R68474	139217	ESTs	4.13	0.77	4	2.12	I	Hs.23294	ESTs		
N51278	283023	Chemokine receptor-like 1	4.54	0.88	5	2.51	I	Hs.78913	G protein-coupled receptor 13	2836	GPR13
AA447781	813823	Lumican	4.55	0.87	5	5.25	I	Hs.79914	lumican	4060	LUM
H93837	242062	Apolipoprotein B (including Ag(x) antigen)	4.55	0.86	5	3.41	I	Hs.585	apolipoprotein B (including Ag(x) antigen)	338	APOB
AA699876	461327	H.sapiens mRNA for phosphoinositide 3-kinase	4.56	0.82	5	2.92	I	Hs.111334	ferritin, light polypeptide	2512	FTL
AA444051	756595	S100 calcium-binding protein A10 (annexin II ligand, calpactin I, light polypeptide	4.56	0.71	5	3.16	I	Hs.119301	S100 calcium-binding protein A10 (annexin II ligand, calpactin I, light polypeptide (p11))	6281	S100A10
AA521339	826142	Human beta2-chimaerin mRNA, complete cds	4.57	0.88	5	2.43	I	Hs.178502	ESTs		
AA486850	841008	Guanylate binding protein 1, interferon-inducible, 67kD	4.59	0.85	5	2.73	I	Hs.62661	guanylate binding protein 1, interferon-inducible, 67kD	2633	GBP1

Table 1

GenBank ID	Image Clone ID	Clone Name	Cluster location	Max. Correlation	template max	Ratio change	Direction	Unigene	Hs Title	Locus ID	Gene Symbol
R31918	134229	ESTs	4.61	0.79	5	5.00	I	Hs.24477	ESTs		
AA873604	1323448	Human cysteine-rich heart protein (hCRHP) mRNA, complete cds	4.62	0.85	5	2.17	I	Hs.17409	cysteine-rich protein 1 (intestinal)	1396	CRIP1
N78582	300137	Homo sapiens mRNA for AMP-activated protein kinase beta 2 subunit	4.63	0.82	5	2.69	I	Hs.50732	protein kinase, AMP-activated, beta 2 non-catalytic subunit	5565	PRKAB2
AA488676	843098	Homo sapiens neuronal tissue-enriched acidic protein (NAP-22) mRNA, complete cds	4.63	0.95	5	3.35	I	Hs.79516	Homo sapiens neuronal tissue-enriched acidic protein (NAP-22) mRNA, complete cds	10409	
H22826	51582	Human zinc-finger domain-containing protein mRNA, partial cds	4.64	0.92	5	6.46	I	Hs.5978	LIM domain only 7	4008	LMO7
R16764	128785	ESTs	4.64	0.72	5	2.63	I	Hs.19479	ESTs		
AA490172	839991	Collagen, type I, alpha-2	4.65	0.86	5	5.22	I	Hs.179573	collagen, type I, alpha 2	1278	COL1A2
AA443284	783998	AF-9 PROTEIN	4.65	0.91	5	2.67	I	Hs.404	myeloid/lymphoid or mixed-lineage leukemia (trithorax (Drosophila) homolog); translocated to, 3	4300	MLLT3
R22788	130276	ESTs	4.66	0.85	5	2.09	I	Hs.203911	ESTs		
H70775 H70774	214006	H.sapiens H2B/l gene	4.67	0.75	5	2.07	I	Hs.146228 Hs.182278	ESTs H2B histone family, member L		H2BFL
AA486365	840687	Mucin 1, transmembrane	4.67	0.87	5	3.13	I	Hs.89603	mucin 1, transmembrane	4582	MUC1
T64905	66731	Rieger syndrome (solurshin)	4.71	0.92	5	32.26	I	Hs.92282	paired-like homeodomain transcription factor 2	5308	PITX2
N32199	272327	Human melanoma antigen recognized by T-cells (MART-1) mRNA	4.71	0.81	5	5.16	I	Hs.154069	melan-A	2315	MLANA
R93149	197054	ESTs	4.73	0.93	5	2.54	I	Hs.105749	Homo sapiens mRNA for KIAA0553 protein, partial cds		

Table 1

GenBank ID	Image Clone ID	Clone Name	Cluster location	Max. Correlation	template max	Ratio change	Direction	Unigene	Hs Title	Locus ID	Gene Symbol
AA465236	814240	Homo sapiens forkhead protein (FKHRL1) mRNA, complete cds	4.75	0.79	5	2.36	I	Hs.14845	forkhead (Drosophila) homolog (rhabdomyosarcoma) like 1	2309	FKHRL1
T98195								Hs.18252	ESTs		
T98194	121792	ESTs	4.77	0.99	5	4.67	I	Hs.106960	ESTs		
AA443090	809456	Human interferon regulatory factor 7 (humirf7) mRNA, complete cds	4.77	0.94	5	2.13	I	Hs.200390	interferon regulatory factor 7	3665	IRF7
R78735		Human guanine nucleotide exchange factor mRNA,						Hs.186533	ESTs son of sevenless (Drosophila)		
R78734	145001	complete cds	4.79	0.77	5	2.23	I	Hs.21371	homolog 1		SOS1
AA243439	685371	Multiple endocrine neoplasia I	4.80	0.94	5	2.56	I	Hs.24297	multiple endocrine neoplasia I	4221	MEN1
W72051	345626	Homo sapiens mRNA for fatty acid binding protein, complete cds	4.81	0.83	5	2.03	I	Hs.26770	fatty acid binding protein 7, brain	2173	FABP7
R36669	136954	ESTs	4.82	0.93	5	2.00	I	Hs.25119	ESTs		
R76732	143887	ESTs	4.84	0.73	5	2.48	I	Hs.154737	Homo sapiens serine protease mRNA, complete cds		
T96123	120964	ESTs	4.84	0.99	5	5.04	I	Hs.17749	ESTs		
AA486200	842784	Phosphate carrier, mitochondrial	4.86	0.98	5	2.08	I	Hs.78713	phosphate carrier, mitochondrial	5250	PHC
AA856874	1374571	Paired basic amino acid cleaving enzyme (furin, membrane associated receptor protein)	4.91	0.90	5	3.25	I	Hs.59242	paired basic amino acid cleaving enzyme (furin, membrane associated receptor protein)	5045	PACE
R46202	152453	Human iroquois-class homeodomain protein IRX-2a mRNA, complete cds	4.91	0.82	5	2.19	I	Hs.25351	iroquois-class homeodomain protein	10265	IRX-2A
AA486275	842836	LEUKOCYTE ELASTASE INHIBITOR	4.93	0.81	5	3.85	I	Hs.183583	protease inhibitor 2 (anti-elastase), monocyte/neutrophil	1992	ELANH2

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Table 1

GenBank ID	Image Clone ID	Clone Name	Cluster location	Max. Correlation	template max	Ratio change	Direction	Unigene	Hs Title	Locus ID	Gene Symbol
H78386	233583	INTERLEUKIN-1 RECEPTOR, TYPE II PRECURSOR	4.94	0.96	5	2.96	I	Hs.25333	interleukin 1 receptor, type II	7850	IL1R2
W79194	346552	Human symplekin mRNA, complete cds	4.94	0.85	5	2.11	I	Hs.107019	symplekin	9417	SYM
H18938	51293	Human aminoacylase-1 (ACY1) mRNA, complete cds	4.96	0.76	5	2.93	I	Hs.79	aminoacylase 1	95	ACY1
AA012867 AA012866	360885	ADP-ribosylation factor 6	4.96	0.82	5	3.36	I	Hs.89474 Hs.180777	ADP-ribosylation factor 6 ESTs	382	ARF6
AA485141	815774	Human Src-like adapter protein mRNA, complete cds	4.97	0.95	5	5.26	I	Hs.75367	Human Src-like adapter protein mRNA, complete cds		
AA668178	852829	Homo sapiens importin-alpha homolog (SRP1gamma) mRNA, complete cds	4.99	0.72	5	2.44	I	Hs.3886	karyopherin alpha 3 (importin alpha 4)	3839	KPNA3
R43778	35077	Human APEG-1 mRNA, complete cds	4.99	0.87	5	2.05	I	Hs.21639	nuclear protein, marker for differentiated aortic smooth muscle and down-regulated with vascular injury	10290	APEG1
AA047257	488956	Human RNA binding protein Etr-3 mRNA, complete cds	5.04	0.79	5	2.47	I	Hs.82321	mab-21 (C. elegans)-like 1	4081	MAB21L1
R42581	31143	Human acidic ribosomal phosphoprotein P0 mRNA, complete cds	5.06	0.81	5	2.09	I	Hs.73742	ribosomal protein, large, P0	6175	RPLP0
AA478436 AA402352	741067	Human SWI/SNF complex 60 KDa subunit (BAF60b) mRNA, complete cds	5.07	0.85	5	2.39	I	Hs.64264 Hs.204365	ESTs SWI/SNF related, matrix associated, actin dependent regulator of chromatin, subfamily d, member 2		SMARCD2

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GenBank ID	Image Clone ID	Clone Name	Cluster location	Max. Correlation	template max	Ratio change	Direction	Unigene	Hs Title	Locus ID	Gene Symbol
AA292226 AA292410	725877	Clusterin (complement lysis inhibitor; testosterone-repressed prostate message 2; apolipoprotein J)	5.07	0.77	5	2.36	I	Hs.204412 Hs.75106	Creatine transporter [human, brainstem/spinal cord, mRNA, 2283 nt] clusterin (complement lysis inhibitor, SP-40,40, sulfated glycoprotein 2, testosterone-repressed prostate message 2, apolipoprotein J)		CLU
W72697	345858	Human cisplatin resistance associated alpha protein (hCRA alpha) mRNA, complete cds	5.08	0.75	5	2.08	I	Hs.166066	Human cisplatin resistance associated beta protein (hCRA beta) mRNA, complete cds		
R45640	35828	Diphtheria toxin receptor (heparin-binding epidermal growth factor-like growth factor)	5.09	0.89	5	3.16	I	Hs.799	diphtheria toxin receptor (heparin-binding epidermal growth factor-like growth factor)	1839	DTR
R43766	34849	Eukaryotic translation elongation factor 2	5.12	0.90	5	2.37	I	Hs.75309	eukaryotic translation elongation factor 2	1938	EEF2
AA706987	431397	H.sapiens mRNA for UDP-GalNAc:polypeptide N-acetylgalactosaminyltransferase (T1)	5.21	0.74	5	2.12	I	Hs.80120	UDP-N-acetyl-alpha-D-galactosamine:polypeptide N-acetylgalactosaminyltransferase 1 (GalNAc-T1)	2589	GALNT1
H55953	203469	ESTs	5.52	0.71	6	2.04	I	Hs.12329	ESTs		
AA292676	713782	Human metargidin precursor mRNA, complete cds	5.63	0.88	6	2.71	I	Hs.92208	a disintegrin and metalloproteinase domain 15 (metargidin)	8751	ADAM15
N70280 W04206	296754	ESTs	5.70	0.88	6	2.40	I	Hs.204700 Hs.49111	ESTs ESTs		
AA495790 AA495846	768370	TRANSFORMING PROTEIN RHOB	5.72	0.79	6	2.55	I	Hs.204354 Hs.93468	ras homolog gene family, member B forkhead (Drosophila)-like 7	388	ARHB FKHL7
N53061	246546	ESTs	5.76	0.77	6	2.05	I	Hs.77365	ESTs		

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T67271	66686	UBIQUINOL-CYTOCHROME C REDUCTASE COMPLEX SUBUNIT VI REQUIRING PROTEIN	5.84	0.78	6	2.50	I	Hs.29797	DNA segment on chromosome X (unique) 648 expressed sequence	8264	DXS648E
AA082943	547058	H.sapiens mRNA for cyclin G1	5.93	0.77	6	2.27	I	Hs.79101	cyclin G1	900	CCNG1
R78558	144924	ESTs	5.95	0.94	6	2.05	I	Hs.7446	Human DNA sequence from PAC 487J7 on chromosome 6q21-22.1. Contains an unknown gene coding for three alternative mRNAs. Contains ESTs, STSs, a BAC end-sequence (GSS) and a CA repeat		
AA418020 AA418118	767475	Human cartilage-specific homeodomain protein Cart-1 mRNA, complete cds	5.98	0.81	6	2.18	I	Hs.27495 Hs.41683	ESTs Human cartilage-specific homeodomain protein Cart-1 mRNA, complete cds		
AA158991	591281	H.sapiens lrp mRNA	6.19	0.87	6	2.01	I	Hs.80680	lung resistance-related protein	9961	LRP
AA922705	1474337	Glycogen phosphorylase B (brain form)	6.20	0.87	6	2.15	I	Hs.75658	phosphorylase, glycogen; brain	5834	PYGB
AA489647	823691	Homo sapiens cyclin G2 mRNA, complete cds	6.27	0.77	6	2.38	I	Hs.79069	cyclin G2	901	CCNG2
AA464595	812955	Human malignant melanoma metastasis-suppressor (KiSS-1) gene, mRNA, complete cds	6.91	0.72	7	2.20	D	Hs.95008	KiSS-1 metastasis-suppressor	3814	KISS1
AA894557	1416782	Creatine kinase B	6.95	0.89	7	2.18	D	Hs.173724	creatine kinase, brain	1152	CKB
H73501	232670	ESTs	6.96	0.94	7	2.12	D	Hs.82389	ESTs		
H44127	183602	KERATIN, TYPE I CYTOSKELETAL 14	6.97	0.80	7	2.12	D	Hs.117729	keratin 14 (epidermolysis bullosa simplex, Dowling-Meara, Koebner)	3861	KRT14

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GenBank ID	Image Clone ID	Clone Name	Cluster location	Max. Correlation	template max	Ratio change	Direction	Unigene	Hs Title	Locus ID	Gene Symbol
AA709271	1343468	Human neural cell adhesion protein (NCAM21) mRNA, complete cds	7.07	0.90	7	2.23	D	Hs.177691	neural cell adhesion molecule 2	4685	NCAM2
N77107	289818	Human methylmalonate semialdehyde dehydrogenase gene, complete cds	7.08	0.70	7	3.00	D	Hs.170008	methylmalonate-semialdehyde dehydrogenase	4329	MMSDH
AA040742	486186	Homo sapiens poly(A) binding protein II (PABP2) gene, complete cds	7.10	0.89	7	2.31	D	Hs.117176	poly(A)-binding protein-2	8106	PABP2
AA478543	784772	GRAVIN	7.13	0.72	7	2.54	D	Hs.788	kinase scaffold protein gravin	9590	GRAVIN
AA115076	491565	Human msg1-related gene 1 (mrg1) mRNA, complete cds	7.14	0.79	7	2.07	D	Hs.82071	Cbp/p300-interacting transactivator, with Glu/Asp-rich carboxy-terminal domain, 2	10370	CITED2
AA598517	897781	Keratin 8	7.20	0.76	7	2.23	D	Hs.78271	keratin 8	3856	KRT8
AA873577	1472150	ATP synthase, H+ transporting, mitochondrial F1 complex, O subunit (oligomycin sensitivity conferring protein)	7.29	0.74	7	2.36	D	Hs.76572	ATP synthase, H+ transporting, mitochondrial F1 complex, O subunit (oligomycin sensitivity conferring protein)	539	ATP5O
AA625655	745343	Regenerating islet-derived 1 alpha (pancreatic stone protein, pancreatic thread	7.32	0.79	7	2.01	D	Hs.1032	regenerating islet-derived 1 alpha (pancreatic stone protein, pancreatic thread protein)	5967	REG1A
AA454098	788256	MITOTIC KINESIN-LIKE PROTEIN-1	7.33	0.83	7	4.43	D	Hs.196172	ESTs		
AA868008	1461138	H.sapiens H4/g gene for H4 histone	7.36	0.72	7	4.73	D	Hs.46423	H4 histone family, member G	8364	H4FG
AA400450	742798	Human mitotic centromere-associated kinesin mRNA, complete cds	7.36	0.81	7	2.48	D	Hs.69360	Homo sapiens mitotic centromere-associated kinesin mRNA, complete cds		

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H72018	214965	Human gamma-glutamyl transpeptidase (GGT) protein mRNA, complete cds	7.42	0.74	7	2.44	D	Hs.135	gamma-glutamyltransferase 1	2678	GGT1
AA488324	842968	Homo sapiens mitotic checkpoint kinase Mad3L (MAD3L) mRNA, complete cds	7.44	0.74	7	3.28	D	Hs.36708	budding uninhibited by benzimidazoles 1 (yeast homolog), beta	701	BUB1B
AA133191	490778	Homo sapiens mRNA for low molecular mass ubiquinone-binding protein, complete cds	7.45	0.88	7	2.30	D	Hs.3709	Homo sapiens mRNA for low molecular mass ubiquinone-binding protein, complete cds		
AA486403	842863	Human mRNA for RTP, complete cds	7.89	0.85	8	2.66	D	Hs.75789	differentiation-related gene 1 (nickel-specific induction protein)	10397	RTP
W95001	415102	Cell division cycle 25C	7.91	0.76	8	2.66	D	Hs.656	cell division cycle 25C	995	CDC25C
AA504625	825606	Human kinesin-like spindle protein HKSP (HKSP) mRNA, complete cds	7.99	0.85	8	2.34	D	Hs.8878	kinesin-like 1	3832	KNSL1
R92435	196303	ESTs	7.99	0.86	8	4.05	D	Hs.34584	ESTs		
AA136707	490995	Homo sapiens lysyl hydroxylase isoform 2 (PLOD2) mRNA, complete cds	8.00	0.93	8	2.51	D	Hs.41270 Hs.41270	procollagen-lysine, 2-oxoglutarate 5-dioxygenase (lysine hydroxylase) 2 procollagen-lysine, 2-oxoglutarate 5-dioxygenase (lysine hydroxylase) 2		PLOD2 PLOD2
AA136566	564803	Human putative M phase phosphoprotein 2 (MPP2) mRNA, complete cds	8.03	0.92	8	2.46	D	Hs.239	forkhead (Drosophila)-like 16	2305	FKHL16
N69491 N90985	292933	Human mRNA for kinesin-related protein, partial cds	8.05	0.95	8	2.05	D	Hs.205205 Hs.20830	ESTs kinesin-like 2		KNSL2
AA487267	841507	PULMONARY SURFACTANT-ASSOCIATED PROTEIN A PRECURSOR	8.06	0.78	8	2.49	D	Hs.177582	surfactant, pulmonary-associated protein A1	6435	SFTPA1
W93379	415089	H.sapiens nek2 mRNA for protein kinase	8.06	1.00	8	4.51	D	Hs.153704	NIMA (never in mitosis gene a)-related kinase	4751	NEK2

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AA449336	785707	Homo sapiens protein regulating cytokinesis 1 (PRC1) mRNA, complete cds	8.08	0.90	8	4.53	D	Hs.5101	Hs.5101 ESTs, Highly similar to protein regulating cytokinesis 1		
AA894577	1492304	Homo sapiens mRNA for nucleolar protein hNop56	8.09	0.96	8	2.09	D	Hs.5092	Homo sapiens mRNA for nucleolar protein hNop56	10528	
AA669443	884867	Eukaryotic translation initiation factor 5 (eIF5)	8.10	0.98	8	2.66	D	Hs.184242	eukaryotic translation initiation factor 5	1983	EIF5
AA446120	774446	ADRENOMEDULLIN PRECURSOR	8.10	0.86	8	3.44	D	Hs.394	adrenomedullin	133	ADM
AA446462	781047	Homo sapiens mitotic checkpoint protein kinase (BUB1) mRNA, complete cds	8.11	0.97	8	4.14	D	Hs.98658	budding uninhibited by benzimidazoles 1 (yeast homolog)	699	BUB1
AA485427	811046	Human mRNA for ESP1/CRP2, complete cds	8.11	0.87	8	2.21	D	Hs.70327	cysteine-rich protein 2	1397	CRIP2
AA411850	727526	Centromere protein E (312kD)	8.11	0.89	8	2.08	D	Hs.75573	centromere protein E (312kD)	1062	CENPE
R06900	126650	ESTs	8.12	0.96	8	3.74	D	Hs.132959	ESTs		
H77486	233274	ESTs	8.13	0.86	8	2.01	D	Hs.64691	Homo sapiens mRNA for KIAA0483 protein, partial cds		
AA464908	838802	Procollagen-proline, 2-oxoglutarate 4-dioxygenase (proline 4-hydroxylase), alpha polypeptide	8.14	0.98	8	2.51	D	Hs.76768	procollagen-proline, 2-oxoglutarate 4-dioxygenase (proline 4-hydroxylase), alpha polypeptide 1	5033	P4HA1
H73968 H73329	232837	ESTs	8.16	0.79	8	7.90	D	Hs.173318 Hs.189975	ESTs Preferentially expressed in colorectal cancer ESTs		FLS353
H08564	45544	SM22-ALPHA HOMOLOG	8.17	0.91	8	2.09	D	Hs.75725	transgelin 2	8407	TAGLN2
N50834	280934	Human mevalonate pyrophosphate decarboxylase (MPD) mRNA, complete cds	8.17	0.84	8	2.11	D	Hs.3828	mevalonate (diphospho) decarboxylase	4597	MVD

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GenBank ID	Image Clone ID	Clone Name	Cluster location	Max. Correlation	template max	Ratio change	Direction	Unigene	Hs Title	Locus ID	Gene Symbol
N71003	294487	Homo sapiens nuclear antigen H731-like protein mRNA, complete cds	8.17	0.81	8	2.85	D	Hs.100407	Homo sapiens nuclear antigen H731-like protein mRNA, complete cds		
AA460927	795936	H.sapiens mRNA for translin	8.18	0.94	8	3.59	D	Hs.75066	translin	7247	TSN
R70488 R70578	141818	U1 SMALL NUCLEAR RIBONUCLEOPROTEIN A	8.18	0.81	8	2.05	D	Hs.16001 Hs.173255	sema domain, immunoglobulin domain (Ig), short basic domain, secreted, 3E small nuclear ribonucleoprotein polypeptide A	9723	SEMA3E SNRPA
R22949	131316	ESTs	8.19	0.91	8	4.01	D	Hs.23448	ESTs NO HOMOLOGY HITS		
AA284072	700792	Human protein phosphatase (KAP1) mRNA, complete cds	8.19	0.98	8	5.07	D	Hs.84113	cyclin-dependent kinase inhibitor 3 (CDK2-associated dual specificity phosphatase)	1033	CDKN3
AA663792	969748	Methylmalonyl Coenzyme A mutase	8.19	0.74	8	3.17	D	Hs.171811	adenylate kinase 2	204	AK2
W55964	340558	Homo sapiens Arp2/3 protein complex subunit p16-Arc (ARC16) mRNA, complete cds	8.20	0.72	8	2.03	D	Hs.82425	Arp2/3 protein complex subunit p16	10092	ARC16
R45059 R19956	34778	Vascular endothelial growth factor	8.20	0.88	8	2.74	D	Hs.73793 Hs.206658	ESTs vascular endothelial growth factor		VEGF
R40897	28469	Human succinyl CoA:3-oxoacid CoA transferase precursor (OXCT) mRNA, complete cds	8.20	0.92	8	2.13	D	Hs.177584	3-oxoacid CoA transferase	5019	OXCT
AA454208	795498	PROTEIN PHPS1-2	8.23	0.83	8	2.50	D	Hs.99364	Human pHS1-2 mRNA with ORF homologous to membrane receptor proteins		
N69286 N94106	293569	ESTs	8.23	0.95	8	2.55	D	Hs.16769 Hs.136005	ESTs ESTs		

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H11346		Human pyrroline-5-carboxylate dehydrogenase (P5CDh) mRNA, long form, complete						Hs.194828	Human pyrroline-5-carboxylate dehydrogenase (P5CDh) mRNA, short form, complete cds		
H11369	47853	cds	8.25	0.84	8	2.57	D	Hs.93842	ESTs		
R96998	200402	ESTs	8.25	0.95	8	2.72	D	Hs.70704	ESTs		
H11346		Human pyrroline-5-carboxylate dehydrogenase (P5CDh) mRNA, long form, complete						Hs.194828	Human pyrroline-5-carboxylate dehydrogenase (P5CDh) mRNA, short form, complete cds		
H11369	47853	cds	8.25	0.80	8	2.25	D	Hs.93842	ESTs		
AA663058	852520	Ubiquinol-cytochrome c reductase core protein II	8.25	0.86	8	2.11	D	Hs.75792	hemoglobin, alpha 1	3039	HBA1
AA504461	825295	LOW-DENSITY LIPOPROTEIN RECEPTOR PRECURSOR	8.26	0.81	8	2.72	D	Hs.153468	low density lipoprotein receptor (familial hypercholesterolemia)	3949	LDLR
R38433	26184	Human mRNA for platelet-type phosphofructokinase, complete cds	8.26	0.74	8	2.76	D	Hs.99910	phosphofructokinase, platelet	5214	PFKP
AA862465	1456160	Zinc-alpha-2-glycoprotein 1	8.26	0.78	8	5.48	D	Hs.71	alpha-2-glycoprotein 1, zinc	563	AZGP1
AA136710	491001	LACTOYLGLUTATHIONE LYASE	8.29	0.71	8	2.38	D	Hs.75207	Human mRNA for lactoyl glutathione lyase		
AA426374		Homo sapiens (clone ch13lambda7) alpha-tubulin						Hs.98102	tubulin, alpha 2		
AA436990	757489	mRNA, complete cds	8.30	0.93	8	2.48	D	Hs.19193	ESTs	7278	TUBA2
W35378	321661	Protein phosphatase 2A, regulatory subunit B' alpha-1	8.30	0.83	8	2.02	D	Hs.171734	Human mRNA for KIAA0044 gene, partial cds		
AA146655	592359	Human kidney mRNA for putative membrane protein with histidine rich charge clusters, complete cds	8.32	0.70	8	2.30	D	Hs.66776	Homo sapiens mRNA for membrane protein with histidine rich charge clusters, complete cds		
H21042	51448	Activating transcription factor 3	8.32	0.75	8	2.89	D	Hs.460	activating transcription factor 3	467	ATF3

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AA291732	725284	Phosphorylase kinase, gamma 2 (testis)	8.32	0.72	8	2.49	D	Hs.196177	ESTs		
H09614	46182	CTP synthetase	8.32	0.83	8	2.79	D	Hs.204665	CTP synthase	1503	CTPS
W49667	324891	Homo sapiens putative fatty acid desaturase MLD mRNA, complete cds	8.33	0.82	8	3.72	D	Hs.185973	membrane fatty acid (lipid) desaturase	8560	MLD
AA485377	811015	P55-C-FOS PROTO-ONCOGENE PROTEIN	8.33	0.83	8	4.80	D	Hs.25647	v-fos FBJ murine osteosarcoma viral oncogene homolog	2353	FOS
R46821	36393	T-COMPLEX PROTEIN 1, ALPHA SUBUNIT	8.34	0.70	8	2.40	D	Hs.4112	acetyl-Coenzyme A acetyltransferase 2 (acetoacetyl Coenzyme A thiolase)	39	ACAT2
H59663	207288	Homo sapiens insulin induced protein 1 (INSIG1) gene, complete cds	8.35	0.87	8	8.07	D	Hs.56205	insulin induced gene 1	3638	INSIG1
AA621218	744417	Carnitine acetyltransferase	8.35	0.83	8	2.37	D	Hs.12068	carnitine acetyltransferase	1384	CRAT
AA447748	813648	Dihydrolipoamide dehydrogenase (E3 component of pyruvate dehydrogenase complex, 2-oxo-glutarate complex, branched chain keto acid dehydrogenase complex)	8.37	0.83	8	2.13	D	Hs.74635	dihydrolipoamide dehydrogenase (E3 component of pyruvate dehydrogenase complex, 2-oxo-glutarate complex, branched chain keto acid dehydrogenase complex)	1738	DLD
AA626698	745138	Human alpha-tubulin isotype H2-alpha gene, last exon	8.38	0.84	8	2.33	D	Hs.98102	tubulin, alpha 2	7278	TUBA2
AA180742	612274	TUBULIN ALPHA-4 CHAIN	8.39	0.78	8	2.08	D	Hs.75318	tubulin, alpha 1 (testis specific)	7277	TUBA1
AA450360	785616	Signal sequence receptor, alpha	8.40	0.79	8	2.65	D	Hs.205072	signal sequence receptor, alpha (translocon-associated protein alpha)	6745	SSR1
H73354	214614	ESTs	8.42	0.75	8	2.57	D	Hs.205626	sorting nexin 4	8723	SNX4
AA680322	869538	Human NADH:ubiquinone oxidoreductase MLRQ subunit mRNA, complete cds	8.91	0.96	9	2.09	D	Hs.108661	NADH dehydrogenase (ubiquinone) 1 alpha subcomplex, 4 (9kD, MLRQ)	4697	NDUFA4

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T47443	71101	Homo sapiens endothelial cell protein C/APC receptor (EPCR) mRNA, complete cds	8.91	0.92	9	2.08	D	Hs.82353	Homo sapiens endothelial cell protein C/APC receptor (EPCR) mRNA, complete cds	10544	
H59231	204257	Human metalloprotease/disintegrin/cysteine-rich protein precursor (MDC9) mRNA, complete cds	8.94	0.89	9	3.23	D	Hs.2442	a disintegrin and metalloproteinase domain 9 (meltrin gamma)	8754	ADAM9
AA663310	853368	Thymidylate synthase	8.95	0.89	9	2.40	D	Hs.196351	Homo sapiens gene for thymidylate synthase, exons 1, 2, 3, 4, 5, 6, 7, complete cds		
AA430504	769921	Human cyclin-selective ubiquitin carrier protein mRNA, complete cds	8.95	0.97	9	5.22	D	Hs.93002	Human cyclin-selective ubiquitin carrier protein mRNA, complete cds ubiquitin carrier protein E2-C 20		UBCH10
W04674 W31775	320509	Homo sapiens mRNA for cytochrome b5, partial cds	8.96	0.83	9	2.60	D	Hs.31086 Hs.206068	ESTs Homo sapiens mRNA for cytochrome b5, partial cds		
H84871 H85277	249603	Homo sapiens DCHT mRNA, complete cds	8.96	0.94	9	3.11	D	Hs.199263 Hs.206211	ESTs ESTs		
H58873	207358	Human (HepG2) glucose transporter gene mRNA, complete cds	8.96	0.99	9	3.47	D	Hs.169902	Human (HepG2) glucose transporter gene mRNA, complete cds		
AA701455	435076	Human CENP-F kinetochore protein mRNA, complete cds	8.96	0.90	9	5.16	D	Hs.77204	centromere protein F (400kD)	1063	CENPF
H94949	242578	Human tastin mRNA, complete cds	8.97	0.88	9	2.16	D	Hs.171955	trophinin-assisting protein (tastin)	10024	TASTIN
R63929 R64020	139705	ESTs	8.97	0.89	9	2.38	D	Hs.28465 Hs.206257	ESTs ESTs		
N75581	299360	Human FUSE binding protein mRNA, complete cds	8.97	0.76	9	2.01	D	Hs.118962	far upstream element binding protein	8880	FUBP

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AA406536	753457	NADH-UBIQUINONE OXIDOREDUCTASE 75 KD SUBUNIT PRECURSOR	8.97	0.86	9	2.69	D	Hs.8248	NADH dehydrogenase (ubiquinone) Fe-S protein 1 (75kD) (NADH-coenzyme Q reductase)	4719	NDUFS1
AA459292	810899	CDC28 protein kinase 1	8.97	0.93	9	2.59	D	Hs.77550	CDC28 protein kinase 1	1163	CKS1
R44288	34355	Human mRNA for calmodulin	8.97	0.70	9	2.54	D	Hs.182278	H2B histone family, member L	8347	H2BFL
N72115	291057	Human CDK6 inhibitor p18 mRNA, complete cds	8.98	0.90	9	2.09	D	Hs.4854	cyclin-dependent kinase inhibitor 2C (p18, inhibits CDK4)	1031	CDKN2C
H05914	43550	Human mRNA for lactate dehydrogenase-A (LDH-A, EC 1.1.1.27)	8.98	0.87	9	3.48	D	Hs.2795	lactate dehydrogenase A	3939	LDHA
AA521466	826211	Programmed cell death 2	8.98	0.89	9	2.06	D	Hs.41639	programmed cell death 2	5134	PDCD2
AA450123	789147	Enolase 2, (gamma, neuronal)	8.98	0.89	9	2.18	D	Hs.196837	H.spaiens 3' mRNA for neurone-specific enolase (EC 4.2.1.11)		
N70382 W05026	295483	ESTs	8.99	0.76	9	2.24	D	Hs.49853 Hs.187111	ESTs ESTs		
AA463510	797016	Succinate dehydrogenase 1, iron sulphur (Ip) subunit	8.99	0.85	9	2.48	D	Hs.64	succinate dehydrogenase complex, subunit B, iron sulfur (Ip)	6390	SDHB
N93924	309288	Replication factor C, 37-kD subunit	8.99	0.84	9	2.38	D	Hs.35120	replication factor C (activator 1) 4 (37kD)	5984	RFC4
H08820	44975	Human homolog of yeast IPP isomerase	8.99	0.98	9	2.60	D	Hs.76038	isopentenyl-diphosphate delta isomerase	3422	IDI1
W86653	416833	Human 54 kDa progesterone receptor-associated immunophilin FKBP54 mRNA, partial cds	9.00	0.96	9	5.84	D	Hs.7557	FK506-binding protein 5	2289	FKBP5
AA497029	897567	L-LACTATE DEHYDROGENASE M CHAIN	9.00	0.78	9	3.08	D	Hs.2795	lactate dehydrogenase A	3939	LDHA
AA456621	809588	Homo sapiens human gamma-glutamyl hydrolase (hGH) mRNA, complete cds	9.00	0.73	9	4.33	D	Hs.78619	gamma-glutamyl hydrolase (conjugase, folylpolygammaglutamyl hydrolase)	8836	GGH

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AA404486	772304	Adenine nucleotide translocator 2 (fibroblast)	9.00	0.98	9	2.53	D	Hs.79172	adenine nucleotide translocator 2 (fibroblast)	292	ANT2
W45531 W45393	323404	ESTs	9.00	0.93	9	2.47	D	Hs.94642 Hs.55888	ESTs Human mRNA for ATF-a transcription factor		
AA458483 AA442991	809603	Prothymosin alpha	9.00	0.75	9	2.35	D	Hs.44222 Hs.182371	ESTs prothymosin, alpha (gene sequence 28)		PTMA
N67038	295986	H.sapiens mRNA for phenylalkylamine binding protein	9.00	0.89	9	2.51	D	Hs.75105	H.sapiens mRNA for phenylalkylamine binding protein		
AA403295	725188	MALATE DEHYDROGENASE, CYTOPLASMIC	9.00	0.88	9	2.54	D	Hs.75375	malate dehydrogenase 1, NAD (soluble)	4190	MDH1
AA629262	744047	Human pLK mRNA, complete cds	9.01	0.85	9	2.37	D	Hs.77597	polo (Drosophia)-like kinase	5347	PLK
AA446839	783697	Homo sapiens E1B 19K/Bcl-2-binding protein Nip3 mRNA, nuclear gene encoding mitochondrial protein, complete cds	9.01	0.75	9	2.86	D	Hs.79428	BCL2/adenovirus E1B 19kD-interacting protein 3	664	BNIP3
AA480995	814615	NAD-DEPENDENT METHYLENETETRAHYDROFOLATE DEHYDROGENASE	9.02	0.85	9	2.67	D	Hs.154672	Human mRNA for NAD-dependent methylene tetrahydrofolate dehydrogenase cyclohydrolase (EC 1.5.1.15)		
AA481076	814701	Homo sapiens mitotic feedback control protein Madp2 homolog mRNA, complete cds	9.02	0.96	9	5.45	D	Hs.79078	MAD2 (mitotic arrest deficient, yeast, homolog)-like 1	4085	MAD2L1
AA598974	898286	Cell division cycle 2, G1 to S and G2 to M	9.02	0.95	9	8.47	D	Hs.206503 Hs.184572	Homo sapiens mRNA for CDC2 delta T, complete cds cell division cycle 2, G1 to S and G2 to M		CDC2 CDC2

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AA608568	950690	Cyclin A	9.02	0.92	9	2.82	D	Hs.85137	cyclin A2	890	CCNA2
T67057	66564	D-BETA-HYDROXYBUTYRATE DEHYDROGENASE	9.03	0.86	9	2.38	D	Hs.76893	3-hydroxybutyrate dehydrogenase (heart, mitochondrial)	622	BDH
AA677257	454339	Thiopurine S-methyltransferase	9.03	0.77	9	2.08	D	Hs.206376	Human thiopurine methyltransferase (TPMT) gene		
H59204	204214	Human Cdc6-related protein (HsCDC6) mRNA, complete cds	9.03	0.92	9	4.70	D	Hs.69563	CDC18 (cell division cycle 18, S.pombe, homolog)-like	990	CDC18L
AA461467	796646	Ornithine decarboxylase 1	9.04	0.89	9	3.84	D	Hs.75212	ornithine decarboxylase 1	4953	ODC1
R19158	129865	Homo sapiens mRNA for aurora/IPL1-related kinase, complete cds	9.04	0.94	9	4.68	D	Hs.199147 Hs.48915	ESTs serine/threonine kinase 15		STK15
AA482117 AA482441	756401	H.sapiens mRNA for ras-related GTP-binding protein	9.04	0.86	9	2.28	D	Hs.177507 Hs.206376	Ras homolog enriched in brain 2 Human thiopurine methyltransferase (TPMT) gene	6009	RHEB2
T66935	66406	ESTs	9.04	0.96	9	5.12	D	Hs.104859	ESTs Blast hits genomic clone AC005538		
AA706968	451907	H.sapiens mRNA for M-phase phosphoprotein, mpp5	9.04	0.97	9	3.11	D	Hs.42650	Homo sapiens ZW10 interactor Zwint mRNA, complete cds		
AA521243	827144	PUTATIVE 60S RIBOSOMAL PROTEIN	9.04	0.75	9	2.27	D	Hs.75574	KIAA0104 gene product	9801	KIAA0104
R12802	25584	Human cytochrome bc-1 complex core protein II mRNA, complete cds	9.06	0.96	9	2.81	D	Hs.75792	hemoglobin, alpha 1	3039	HBA1
AA025937	365641	DNA primase polypeptide 1 (49kD)	9.07	0.97	9	2.04	D	Hs.82741	primase, polypeptide 1 (49kD)	5557	PRIM1
AA482243	840894	CYTOCHROME C OXIDASE POLYPEPTIDE VIA-LIVER PRECURSOR	9.07	0.87	9	2.51	D	Hs.180714	cytochrome c oxidase subunit VIa polypeptide 1	1337	COX6A1

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AA447588	782679	Homo sapiens dead box, Y isoform (DBY) mRNA, alternative transcript 2, complete cds	9.07	0.78	9	2.30	D	Hs.99120	DEAD/H (Asp-Glu-Ala-Asp/His) box polypeptide, Y chromosome	8653	DBY
AA280832	711768	Homo sapiens UDP-galactose-4-epimerase (GALE) mRNA, complete cds	9.08	0.86	9	3.07	D	Hs.76057	ESTs		
H69335	234237	H.sapiens mRNA for Pirin, isolate 1	9.09	0.92	9	2.29	D	Hs.38842	pirin	8544	PIR
W86653	416833	Human 54 kDa progesterone receptor-associated immunophilin FKBP54 mRNA, partial cds	9.10	0.89	9	3.69	D	Hs.7557	FK506-binding protein 5	2289	FKBP5
AA629558	884718	Human histone stem-loop binding protein (SLBP) mRNA, complete cds	9.11	0.89	9	2.19	D	Hs.75257	H.sapiens mRNA histone RNA hairpin-binding protein		
AA453410	788185	Homo sapiens TRAIL receptor 2 mRNA, complete cds	9.65	0.82	10	2.95	D	Hs.51233	tumor necrosis factor receptor superfamily, member 10b	8795	TNFRSF10B
AA700904	453107	Homo sapiens Porc-PI gene similar to yeast CDC45	9.66	0.85	10	3.05	D	Hs.114311	CDC45 (cell division cycle 45, S.cerevisiae, homolog)-like	8318	CDC45L
H55921	204148	Human insulin-stimulated protein kinase 1 (ISPK-1) mRNA, complete cds	9.66	0.72	10	2.27	D	Hs.173965	ribosomal protein S6 kinase, 90kD, polypeptide 3	6197	RPS6KA3
AA056465	509887	Human 54 kDa protein mRNA, complete cds	9.66	0.84	10	3.93	D	Hs.172207	myeloid/lymphoid or mixed-lineage leukemia (trithorax (Drosophila) homolog); translocated to, 7	4303	MLLT7
N62761	289551	Human fragile X mental retardation protein 1 homolog FXR1 mRNA, complete cds	9.67	0.80	10	2.55	D	Hs.82712	fragile X mental retardation, autosomal homolog 1	8087	FXR1
AA479781	740554	Radixin	9.68	0.84	10	2.28	D	Hs.203914	radixin	5962	RDX

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H68272	211747	ESTs	9.68	0.91	10	2.14	D	Hs.38703	ESTs		
T64893	66728	Ferrochelatase	9.68	0.85	10	6.31	D	Hs.26	ferrochelatase (protoporphyria)	2235	FECH
AA450205	789204	Human mRNA for translocation protein-1, complete cds	9.68	0.90	10	3.51	D	Hs.8146	translocation protein 1	7095	TLOC1
AA018980	362926	Protein kinase, cAMP-dependent, catalytic, beta	9.69	0.91	10	2.24	D	Hs.87773	protein kinase, cAMP-dependent, catalytic, beta	5567	PRKACB
AA775355	878676	ATP-DEPENDENT DNA HELICASE II, 86 KD SUBUNIT	9.71	0.89	10	2.36	D	Hs.84981	X-ray repair complementing defective repair in Chinese hamster cells 5 (double-strand-break rejoining; Ku autoantigen, 80kD)	7520	XRCC5
AA173109 AA173369	611150	Human mitochondrial ATP synthase subunit 9, P3 gene copy, mRNA, nuclear gene encoding mitochondrial protein, complete cds	9.71	0.77	10	2.58	D	Hs.22893 Hs.429	ESTs ATP synthase, H+ transporting, mitochondrial F0 complex, subunit c (subunit 9) isoform 3		ATP5G3
H17504	50506	H.sapiens ERK3 mRNA	9.71	0.80	10	2.21	D	Hs.75465	protein kinase, mitogen-activated 6 (extracellular signal-regulated kinase, p97)	5597	PRKM6
H27646	162775	P68 PROTEIN	9.71	0.73	10	6.00	D	Hs.76053	DEAD/H (Asp-Glu-Ala-Asp/His) box polypeptide 5 (RNA helicase, 68kD)	1655	DDX5
AA400234	743230	T3 receptor-associating cofactor-1 [human, fetal liver, mRNA, 2930 nt]	9.73	0.71	10	2.99	D	Hs.120980	T3 receptor-associating cofactor-1 [human, fetal liver, mRNA, 2930 nt]		smrt
AA191548 AA191245	626716	Human RNA polymerase II elongation factor ELL2, complete cds	9.74	0.89	10	3.29	D	Hs.61933 Hs.173334	ESTs Human RNA polymerase II elongation factor ELL2, complete cds		
H11622	47833	Homo sapiens endothelin-1 (EDN1)	9.75	0.83	10	3.40	D	Hs.50144	Tubulin, alpha, brain-specific	7846	TUBA3

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AA460838	796278	H.sapiens mRNA for basic transcription factor 2, 34 kD subunit	9.76	0.90	10	2.53	D	Hs.90304	general transcription factor IIH, polypeptide 3 (34kD subunit)	2967	GTF2H3
R12473	128243	Adenosine kinase	9.76	0.89	10	2.16	D	Hs.94382	adenosine kinase	132	ADK
T70056	80946	Ribonuclease L (2',5'-oligoisoadenylate synthetase-dependent) inhibitor	9.77	0.79	10	2.27	D	Hs.12013	ribonuclease L (2',5'-oligoisoadenylate synthetase-dependent) inhibitor	6059	RNASELI
R82300	149013	S-adenosylmethionine decarboxylase 1	9.77	0.92	10	3.97	D	Hs.205111	S-adenosylmethionine decarboxylase 1	262	AMD1
AA071486	531319	Homo sapiens protein kinase mRNA, complete cds	9.77	0.87	10	7.01	D	Hs.180655	serine/threonine kinase 12	9212	STK12
AA599116	950482	Small nuclear ribonucleoprotein polypeptides	9.78	0.92	10	2.38	D	Hs.83753	small nuclear ribonucleoprotein polypeptides B and B1	6628	SNRPB
AA599120	950473	Homo sapiens BAF57 (BAF57) gene, complete cds	9.78	0.92	10	2.17	D	Hs.3404	SWI/SNF related, matrix associated, actin dependent regulator of chromatin, subfamily e, member 1	6605	SMARCE1
AA450265	789182	Proliferating cell nuclear antigen	9.79	0.98	10	3.09	D	Hs.78996	proliferating cell nuclear antigen	5111	PCNA
AA630784	856427	Homo sapiens HPV16 E1 protein binding protein mRNA, complete cds	9.79	0.91	10	5.10	D	Hs.6566	Homo sapiens HPV16 E1 protein binding protein mRNA, complete cds		
AA397813	725454	CDC28 protein kinase 2	9.80	0.94	10	6.18	D	Hs.83758	CDC28 protein kinase 2	1164	CKS2
AA620553	951142	FLAP ENDONUCLEASE-1	9.80	0.92	10	2.92	D	Hs.4756	flap structure-specific endonuclease	2237	FEN1
AA775415	878130	H.sapiens mRNA for SMT3B protein	9.81	0.83	10	2.01	D	Hs.180139	H.sapiens mRNA for SMT3B protein		
H15446	49352	Annexin VII (synexin)	9.82	0.85	10	3.02	D	Hs.78637	annexin VII (synexin)	310	ANX7
W45148	322914	Acid phosphatase 1, soluble	9.82	0.80	10	2.11	D	Hs.75393	acid phosphatase 1, soluble	52	ACP1
AA872341	1472643	40S RIBOSOMAL PROTEIN S15A	9.83	0.87	10	2.63	D	Hs.2953	ribosomal protein S15a	6210	RPS15A
N69694	293727	ESTs	9.83	0.87	10	4.07	D	Hs.49738	ESTs		

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N33274	273546	MULTIFUNCTIONAL PROTEIN ADE2	9.83	0.90	10	3.33	D	Hs.117950	H.sapiens ADE2H1 mRNA showing homologies to SAICAR synthetase and AIR carboxylase of the purine pathway (EC 6.3.2.6, EC 4.1.1.21)		
AA404387	772220	Human mRNA for protein disulfide isomerase-related protein (PDIR), complete cds	9.83	0.91	10	2.20	D	Hs.76901	Homo sapiens mRNA for protein disulfide isomerase-related protein (PDIR), complete cds		
R15814	53316	Human malate dehydrogenase (MDHA) mRNA, complete cds	9.84	0.83	10	3.28	D	Hs.75375	malate dehydrogenase 1, NAD (soluble)	4190	MDH1
R60317	42059	Human dihydrolipoamide dehydrogenase mRNA, complete cds	9.84	0.91	10	3.44	D	Hs.74635	dihydrolipoamide dehydrogenase (E3 component of pyruvate dehydrogenase complex, 2-oxo-glutarate complex, branched chain keto acid dehydrogenase complex)	1738	DLD
AA458957	810854	Human RNaseP protein p30 (RPP30) mRNA, complete cds	9.84	0.85	10	2.35	D	Hs.139120	Human RNaseP protein p30 (RPP30) mRNA, complete cds		
AA424950	768260	RETINOBLASTOMA BINDING PROTEIN 3	9.85	0.86	10	2.52	D	Hs.96055	E2F transcription factor 1	1869	E2F1
AA186327	626531	Homo sapiens RRM RNA binding protein Gry-rbp (GRY-RBP) mRNA, complete cds	9.85	0.89	10	2.68	D	Hs.155489	NS1-associated protein 1	10492	NSAP1
AA035384	471598	Homo sapiens mRNA for small subunit of cytochrome b in succinate dehydrogenase complex, complete cds	9.85	0.87	10	2.08	D	Hs.168289	succinate dehydrogenase complex, subunit D, integral membrane protein	6392	SDHD
H06853	44537	Unknown EST	9.85	0.98	10	2.67	D	Hs.83753	small nuclear ribonucleoprotein polypeptides B and B1	6628	SNRPB

Table 1

GenBank ID	Image Clone ID	Clone Name	Cluster location	Max. Correlation	template max	Ratio change	Direction	Unigene	Hs Title	Locus ID	Gene Symbol
AA425908	769603	Human arfaptin 2, putative target protein of ADP-ribosylation factor, mRNA, complete cds	9.85	0.80	10	2.25	D	Hs.75139	ESTs		
AA047567	376785	Homo sapiens mRNA for putative progesterone binding protein	9.85	0.70	10	2.31	D	Hs.9071	Homo sapiens mRNA for putative progesterone binding protein	10424	
AA426227	760344	Uridine monophosphate synthetase (orotate phosphoribosyl transferase and orotidine-5'-decarboxylase)	9.86	0.82	10	2.10	D	Hs.2057	uridine monophosphate synthetase (orotate phosphoribosyl transferase and orotidine-5'-decarboxylase)	7372	UMPS
AA485626	840364	S-adenosylhomocysteine hydrolase	9.86	0.92	10	2.04	D	Hs.172673	S-adenosylhomocysteine hydrolase	191	AHCY
AA633549	856489	RIBONUCLEOSIDE-DIPHOSPHATE REDUCTASE M1 CHAIN	9.86	0.89	10	2.14	D	Hs.2934	ribonucleotide reductase M1 polypeptide	6240	RRM1
AA122272	490772	U2 SMALL NUCLEAR RIBONUCLEOPROTEIN A'	9.86	0.89	10	2.02	D	Hs.80506	small nuclear ribonucleoprotein polypeptide A'	6627	SNRPA1
H09590	46171	Human mRNA for eukaryotic initiation factor 4AI	9.86	0.97	10	2.72	D	Hs.159554	Human clone 23933 mRNA sequence		
AA664155	855487	Human putative 32kDa heart protein PHP32 mRNA, complete cds	9.87	0.93	10	3.49	D	Hs.75811	N-acylsphingosine amidohydrolase (acid ceramidase)	427	ASAH
N50880	281003	T cell receptor gamma chain	9.87	0.82	10	5.05	D	Hs.112259	T-cell receptor, gamma cluster	6965	TCRG
AA047319	509495	PROTEASOME IOTA CHAIN	9.87	0.90	10	2.32	D	Hs.74077	proteasome (prosome, macropain) subunit, alpha type, 6	5687	PSMA6
H05893	43231	Human mRNA for 26S proteasome subunit p97	9.88	0.75	10	2.48	D	Hs.74619	proteasome (prosome, macropain) 26S subunit, non-ATPase, 2	5708	PSMD2
AA045529	487348	Homo sapiens dynamin-like protein mRNA, complete cds	9.88	0.95	10	2.97	D	Hs.180628	dynamin-like protein	10059	DYMPLE

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AA598776	898062	Human p55CDC mRNA, complete cds	9.88	0.93	10	3.44	D	Hs.82906	cell division cycle 20, S.cerevisiae homolog	991	CDC20
AA460286	795738	Human G protein gamma-10 subunit mRNA, complete cds	9.89	0.99	10	2.22	D	Hs.79126	guanine nucleotide binding protein 10	2790	GNG10
R37953	24145	Homo sapiens adenylyl cyclase-associated protein (CAP) mRNA, complete cds	9.89	0.84	10	3.18	D	Hs.104125	adenylyl cyclase-associated protein	10487	CAP
R53311	40017	Cytochrome c-1	9.89	0.97	10	3.00	D	Hs.697	cytochrome c-1	1537	CYC1
AA004759	429182	Homo sapiens dolichol monophosphate mannose synthase (DPM1) mRNA, partial cds	9.89	0.90	10	2.01	D	Hs.5085	dolichyl-phosphate mannosyltransferase polypeptide 1, catalytic subunit	8813	DPM1
AA504809	825677	Homo sapiens arsenite translocating ATPase (ASNA1) mRNA, complete cds	9.89	0.79	10	2.51	D	Hs.165439	arsA (bacterial) arsenite transporter, ATP-binding, homolog 1	439	ASNA1
AA422058 AA422139	755239	H.sapiens mRNA for D1075-like gene	9.90	0.93	10	2.45	D	Hs.42957 Hs.75663	methyltransferase-like 1 non-metastatic cells 2, protein (NM23B) expressed in	4234	METTL1 NME2
AA683050	971367	40S RIBOSOMAL PROTEIN	9.91	0.88	10	2.65	D	Hs.118690	ribosomal protein S8	6202	RPS8
R10947	129146	Homo sapiens mRNA for COX7RP, complete cds	9.92	0.87	10	2.54	D	Hs.30888	cytochrome c oxidase subunit VII-related protein	9167	COX7RP
AA504540	825312	ATP synthase, H+ transporting, mitochondrial	9.92	0.84	10	2.57	D	Hs.73851	ATP synthase, H+ transporting, mitochondrial F0 complex, subunit F6	522	ATP5J
AA702548	384081	Homo sapiens Pig8 (PIG8) mRNA, complete cds	9.92	0.86	10	2.15	D	Hs.8141	etoposide-induced mRNA	9538	PIG8
R37665	26578	Homo sapiens pescadillo mRNA, complete cds	9.92	0.78	10	3.70	D	Hs.13501	Homo sapiens pescadillo mRNA, complete cds		
AA459013	814287	Homo sapiens X-ray repair cross-complementing protein 3 (XRCC3) mRNA, complete cds	9.92	0.70	10	3.02	D	Hs.99742	X-ray repair complementing defective repair in Chinese hamster cells 3	7517	XRCC3

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AA452278	787938	Homo sapiens sodium bicarbonate cotransporter (HNBC1) mRNA, complete cds	9.93	0.92	10	2.16	D	Hs.5462	solute carrier family 4, sodium bicarbonate cotransporter, member 4	8671	SLC4A4
AA669689	857264	MALATE OXIDOREDUCTASE	9.93	0.79	10	2.62	D	Hs.14732	malic enzyme 1, soluble	4199	ME1
AA453750	813675	Human D9 splice variant A mRNA, complete cds	9.93	0.96	10	2.64	D	Hs.37616	Human D9 splice variant B mRNA, complete cds		
R28287								Hs.205702	ESTs		
R28071	134719	ESTs	9.94	0.89	10	3.68	D	Hs.170047	ESTs		
AA188761	626206	DNA polymerase gamma	9.94	0.95	10	2.21	D	Hs.198574	ESTs		
AA251770	684655	26S PROTEASE REGULATORY SUBUNIT 7	9.95	0.87	10	3.20	D	Hs.61153	proteasome (prosome, macropain) 26S subunit, ATPase, 2	5701	PSMC2
R40970								Hs.90797	Homo sapiens clone 23620 mRNA sequence		
R14475	29063	ESTs	9.95	0.90	10	3.14	D	Hs.119007	ESTs		
AA699317	433666	H.sapiens mRNA for testican	9.95	0.98	10	2.44	D	Hs.93029	sparc/osteonectin, cwcv and kazal-like domains proteoglycan (testican)	6695	SPOCK
R60933	42096	Human cytoplasmic chaperonin hTRiC5 mRNA, partial cds	9.96	0.81	10	2.19	D	Hs.1708	TCP1 (t-complex-1) ring complex, polypeptide 5	7203	TRIC5
W51985								Hs.204503	Human pregnancy-specific beta-glycoprotein e mRNA, complete cds		
W52627	325641	Pregnancy specific beta-1 glycoprotein 5	9.96	0.90	10	2.70	D	Hs.206127	pregnancy specific beta-1-glycoprotein 5		PSG5
N55480	246120	Human mRNA for suppressor for yeast mutant, complete cds	9.96	0.89	10	2.37	D	Hs.20521	HMT1 (hnRNP methyltransferase, S. cerevisiae)-like 2	3276	HRMT1L2
T65790	80410	Farnesyl diphosphate synthase (farnesyl pyrophosphate synthetase, dimethylallyltranstransferase, geranyltranstransferase)	9.96	0.81	10	2.12	D	Hs.77393	farnesyl diphosphate synthase (farnesyl pyrophosphate synthetase, dimethylallyltranstransferase, geranyltranstransferase)	2224	FDPS

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AA877347	1160558	6-PYRUVOYL TETRAHYDROBIOPTERIN SYNTHASE	9.97	0.94	10	2.56	D	Hs.366	6-pyruvoyltetrahydropterin synthase	5805	PTS
H07880	45233	Human chaperonin protein (Tcp20) gene complete cds	9.97	0.81	10	3.01	D	Hs.82916	chaperonin containing T-complex subunit 6	908	CCT6
H73928	214884	PROTEIN TRANSPORT PROTEIN SEC61 BETA SUBUNIT	9.97	0.95	10	2.37	D	Hs.77028	Human Sec61-complex beta-subunit mRNA, complete cds		
AA489011	824906	Homo sapiens thyroid receptor interactor (TRIP3) mRNA, 3' end of cds	9.97	0.93	10	2.04	D	Hs.196326	ESTs		
AA465386	814117	Human Gu protein mRNA, partial cds	9.97	0.82	10	4.00	D	Hs.5122	ESTs		
AA465019	810057	DNA-BINDING PROTEIN A	9.97	0.73	10	2.24	D	Hs.1139	cold shock domain protein A	8531	CSDA
AA863149	1455641	Homo sapiens proteasome subunit XAPC7 mRNA, complete cds	9.98	0.98	10	2.33	D	Hs.119502	proteasome (prosome, macropain) subunit, alpha type, 7	5688	PSMA7
AA079059	545403	Homo sapiens thioredoxin-like protein mRNA, complete cds	9.98	0.77	10	2.60	D	Hs.18792	thioredoxin-like, 32kD	9352	TXNL
AA425755	773383	Homo sapiens mRNA for leukemia associated gene 1	9.99	0.82	10	2.38	D	Hs.20149	Homo sapiens mRNA for leukemia associated gene 1	10301	
AA488081	840702	Human selenium donor protein (selD) mRNA, complete cds	9.99	0.81	10	2.70	D	Hs.124027	Human selenium donor protein (selD) mRNA, complete cds		
AA464567	810552	Human B-cell receptor associated protein (hBAP) mRNA, partial cds	9.99	0.90	10	2.78	D	Hs.7771	Human B-cell receptor associated protein (hBAP) mRNA, partial cds		
AA479030	739511	Human kinase Myt1 (Myt1) mRNA, complete cds	10.00	0.95	10	2.71	D	Hs.77783	membrane-associated tyrosine- and threonine-specific cdc2-inhibitory kinase	9088	PKMYT1

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R77517	145503	Human CDK inhibitor p19INK4d mRNA, complete	10.00	0.88	10	2.78	D	Hs.29656	cyclin-dependent kinase inhibitor 2D (p19, inhibits CDK4)	1032	CDKN2D
R32755	135449	Ewing sarcoma breakpoint region 1	10.01	0.78	10	2.03	D	Hs.129953	Ewing sarcoma breakpoint region 1	2130	EWSR1
AA253448	724387	Human mRNA for U1 small nuclear RNP-specific C protein	10.01	0.89	10	2.00	D	Hs.1063	small nuclear ribonucleoprotein polypeptide C	6631	SNRPC
N54768	283315	Phosphoglycerate mutase 2 (muscle)	10.01	0.94	10	3.68	D	Hs.46039	phosphoglycerate mutase 2 (muscle)	5224	PGAM2
AA676970	897177	Phosphoglycerate mutase 1 (brain)	10.01	0.82	10	2.01	D	Hs.181013	phosphoglycerate mutase 1 (brain)	5223	PGAM1
AA679345	866874	Human BTK region clone ftp-3 mRNA	10.01	0.83	10	2.22	D	Hs.75927	Human BTK region clone ftp-3 mRNA		
AA127093	502669	Human transcriptional regulator homolog RPD3 mRNA, complete cds	10.03	0.96	10	2.02	D	Hs.3352	histone deacetylase 2	3066	HDAC2
W47014	324618	MITOCHONDRIAL ELONGATION FACTOR TS PRECURSOR	10.03	0.84	10	2.18	D	Hs.3273	Ts translation elongation factor, mitochondrial	10102	TSFM
AA598510	897774	Human APRT gene for adenine phosphoribosyltransferase	10.04	0.77	10	3.31	D	Hs.28914	adenine phosphoribosyltransferase	353	APRT
T55801	73381	Human TFIIA gamma subunit mRNA, complete cds	10.04	0.85	10	2.44	D	Hs.76362	general transcription factor IIA, 1 (37kD and 19kD subunits)	2957	GTF2A1
R58991	41452	Spermidine/spermine N1-acetyltransferase mRNA, complete cds	10.05	0.76	10	2.37	D	Hs.195207	glutathione S-transferase pi	2950	GSTP1
W44860	321389	Human calmodulin mRNA, complete cds	10.06	0.80	10	2.85	D	Hs.78271	keratin 8	3856	KRT8
AA600217	949971	CAMP-dependent transcription factor ATF-4 (CREB2)	10.06	0.79	10	2.07	D	Hs.181243	activating transcription factor 4 (tax-responsive enhancer element B67)	468	ATF4

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AA449693 AA448967	785778	Homo sapiens spleen mitotic checkpoint BUB3 (BUB3) mRNA, complete cds	10.06	0.87	10	3.03	D	Hs.112318 Hs.40323	Homo sapiens mRNA for 6.2 kd protein BUB3 (budding uninhibited by benzimidazoles 3, yeast) homolog		BUB3 BUB3
H17096	50359	Unknown EST	10.06	0.70	10	2.38	D	Hs.75694	mannose phosphate isomerase	4351	MPI
AA455931	813280	Adenylosuccinate lyase	10.07	0.92	10	2.39	D	Hs.75527	adenylosuccinate lyase	158	ADSL
N95462	308682	Homo sapiens mRNA for putative ABC transporter,	10.07	0.72	10	2.11	D	Hs.153612	Homo sapiens mRNA for putative ABC transporter, partial		
AA113903	531957	Human mRNA for ubiquitin-conjugating enzyme, complete cds	10.09	0.90	10	2.07	D	Hs.78563	ubiquitin-conjugating enzyme E2G 1 (homologous to C. elegans UBC7)	7326	UBE2G1
AA043133	486175	Solute carrier family 16 (monocarboxylic acid transporters), member 1	10.09	0.87	10	2.46	D	Hs.75231	solute carrier family 16 (monocarboxylic acid transporters), member 1	6566	SLC16A1
R53942	40026	Human mitochondrial ADP/ADT translocator mRNA, complete cds	10.09	0.92	10	2.21	D	Hs.2043	adenine nucleotide translocator 1 (skeletal muscle)	291	ANT1
R40767	28823	Human mRNA for clathrin-like protein, complete cds	10.09	0.70	10	2.36	D	Hs.77770	Human mRNA for clathrin-like protein, complete cds		
AA046590	487425	H.sapiens mRNA for centrin gene	10.11	0.87	10	2.60	D	Hs.29463	centrin, EF-hand protein, 3 (CDC31 yeast homolog)	1070	CETN3
R59697 R59807	42880	CELL DIVISION PROTEIN KINASE 8	10.12	0.80	10	2.04	D	Hs.25283 Hs.206758	ESTs cyclin-dependent kinase 8		CDK8
W86139	416316	H.sapiens mRNA for nuclear protein SDK3, partial	10.14	0.83	10	2.03	D	Hs.44499	small EDRK-rich factor 2	10169	SERF2
R43325	32875	Human ionizing radiation resistance conferring protein mRNA, complete	10.15	0.81	10	2.13	D	Hs.159627	Death associated protein 3	7818	DAP3
AA410517 AA411343	753862	Cytoplasmic antiproteinase	10.54	0.80	11	2.46	D	Hs.41072 Hs.539	Cytoplasmic antiproteinase ribosomal protein S29		RPS29

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AA459663	795543	Human antioxidant enzyme AOE37-2 mRNA, complete cds	10.56	0.84	11	2.58	D	Hs.83383	Human antioxidant enzyme AOE37-2 mRNA, complete cds	10549	
H99681	263727	POLYPOSIS LOCUS PROTEIN 1	10.59	0.90	11	2.13	D	Hs.178112	DNA segment, single copy probe LNS-CAI/LNS-CAII (deleted in	7905	D5S346
AA490981	824568	Prostate specific antigen	10.60	0.71	11	2.77	D	Hs.171995	kallikrein 3, (prostate specific antigen)	354	KLK3
AA062814 AA082419	366156	ESTs	10.62	0.75	11	2.34	D	Hs.205357 Hs.177530	ESTs ESTs		
AA878561	1492412	Ubiquitin A-52 residue ribosomal protein fusion product 1	10.64	0.87	11	2.00	D	Hs.119502	proteasome (prosome, macropain) subunit, alpha type, 7	5688	PSMA7
AA487893	840567	TUMOR-ASSOCIATED ANTIGEN L6	10.64	0.94	11	7.10	D	Hs.3337	transmembrane 4 superfamily member 1	4071	TM4SF1
H20856	51666	Human DNA repair helicase (ERCC3) mRNA, complete cds	10.64	0.76	11	2.09	D	Hs.77929	excision repair cross-complementing rodent repair deficiency, complementation group 3 (xeroderma pigmentosum group B complementing)	2071	ERCC3
T52362	72050	H.sapiens mRNA for Icln protein	10.66	0.85	11	2.27	D	Hs.84974	chloride channel, nucleotide-sensitive, 1A	1207	CLNS1A
H16256	47647	Human mRNA for dihydropyrimidinase related protein-3, complete cds	10.68	0.81	11	2.14	D	Hs.74566	dihydropyrimidinase-like 3	1809	DPYSL3
AA456478	809535	Splicing factor, arginine/serine-rich 2	10.69	0.86	11	2.39	D	Hs.73965	splicing factor, arginine/serine-rich 2	6427	SFRS2
AA457717	810725	Human mRNA for proton-ATPase-like protein, complete cds	10.71	0.75	11	2.03	D	Hs.7476	ATPase, H+ transporting, lysosomal (vacuolar proton pump) 21kD	533	ATP6F
AA486233	842825	G1 to S phase transition 1	10.72	0.91	11	2.05	D	Hs.2707	G1 to S phase transition 1	2935	GSPT1

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AA418689	767817	DNA-DIRECTED RNA POLYMERASE II 14.4 KD POLYPEPTIDE	10.72	0.95	11	3.10	D	Hs.46405	H.Sapiens gene for RNA polymerase II subunit 14.4 kD		
T64482	80500	Esterase D/formylglutathione hydrolase	10.72	0.91	11	2.05	D	Hs.82193	esterase D/formylglutathione hydrolase	2098	ESD
AA630794	856454	Antigen identified by monoclonal antibodies 4F2, TRA1.10, TROP4, and T43	10.73	0.77	11	2.09	D	Hs.79748	Homo sapiens clone 24551 mRNA sequence		
AA485734	811150	H.sapiens mRNA for RanGTPase activating protein	10.74	0.90	11	2.16	D	Hs.183800	Ran GTPase activating protein 1	5905	RANGAP1
AA032090 AA034103	470930	Homo sapiens actin-related protein Arp2 (ARP2) mRNA, complete cds	10.75	0.96	11	2.41	D	Hs.27424 Hs.62461	DEAD/H (Asp-Glu-Ala-Asp/His) box polypeptide 11 (S.cerevisiae CHL1-like helicase) ARP2 (actin-related protein 2, yeast) homolog	1663	DDX11 ACTR2
H94262 H94162	242698	ESTs	10.76	0.80	11	2.42	D	Hs.41958 Hs.28669	ESTs ESTs		
T68317	83279	Homo sapiens inner mitochondrial membrane translocase Tim23 (TIM23) mRNA, nuclear gene encoding mitochondrial protein, complete cds	10.76	0.99	11	2.04	D	Hs.11866	Homo sapiens inner mitochondrial membrane translocase Tim23 (TIM23) mRNA, nuclear gene encoding mitochondrial protein, complete cds	10431	
AA629862	884993	Homo sapiens mRNA for smallest subunit of ubiquinol-cytochrome c reductase, complete cds	10.77	0.89	11	2.03	D	Hs.8372	Homo sapiens mRNA for smallest subunit of ubiquinol-cytochrome c reductase, complete cds		
H68838	211275	ORPHAN RECEPTOR TR2	10.77	0.90	11	2.20	D	Hs.108301	TR2 nuclear hormone receptor	7181	TR2

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AA40289 AA401361	741958	Homo sapiens NBMPR-insensitive nucleoside transporter ei (ENT2) mRNA, complete cds	10.82	0.74	11	2.20	D	Hs.32951 Hs.12152	equilibrative nucleoside transporter 2 (hydrophobic nucleolar protein, 36kD) ESTs	3177	ENT2
AA699560	433474	Surfeit 1	10.84	0.98	11	2.02	D	Hs.3196	surfeit 1	6834	SURF1
	196650	ESTs	10.84	0.93	11	2.06	D				
AA431433	782439	Homo sapiens mRNA for ATP synthase subunit e, complete cds	10.85	0.74	11	2.38	D	Hs.85539	Homo sapiens mRNA for ATP synthase subunit e, complete cds		
AA486518	843121	Human nuclear chloride ion channel protein (NCC27) mRNA, complete cds	10.85	0.74	11	2.03	D	Hs.74276	chloride intracellular channel 1	1192	CLIC1
R40634	28410	Human AMP deaminase (AMPD2) mRNA	10.86	0.87	11	2.52	D	Hs.82927	Human AMP deaminase isoform L (AMPD2) mRNA, exons 6-18, partial cds		
AA777192	378502	DNA-DIRECTED RNA POLYMERASE II 14.5 KD POLYPEPTIDE	10.88	0.89	11	2.09	D	Hs.47062	polymerase (RNA) II (DNA directed) polypeptide I (14.5kD)	5438	POLR2I
H74133	214906	ESTs	10.88	0.80	11	2.30	D	Hs.184245	ESTs		
H05919	43241	Human mRNA for eukaryotic initiation factor 4AII	10.89	0.80	11	2.32	D	Hs.173912	eukaryotic translation initiation factor 4A, isoform 2	1974	EIF4A2
H16255	47542	SMALL NUCLEAR RIBONUCLEOPROTEIN SM D1	10.90	0.97	11	2.11	D	Hs.86948	Human autoantigen small nuclear ribonucleoprotein Sm-D mRNA, complete cds		
T84249	109863	Human XMP mRNA, complete cds	10.90	0.81	11	2.82	D	Hs.29191	epithelial membrane protein 2	2013	EMP2
AA454947	814765	H.sapiens mRNA for kinase A anchor protein	10.91	0.99	11	2.21	D	Hs.78921	A kinase anchor protein, 149kD	8165	AKAP149
AA702640	384015	Dopa decarboxylase (aromatic L-amino acid decarboxylase)	10.91	0.70	11	3.94	D	Hs.150403	dopa decarboxylase (aromatic L-amino acid decarboxylase)	1644	DDC

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AA099855	489823	Homo sapiens COX17 mRNA, complete cds	10.92	0.71	11	2.33	D	Hs.16297	human homolog of yeast mitochondrial copper recruitment gene	10063	COX17
T69468	83011	Ribosomal protein S4, Y-linked	10.93	0.87	11	2.10	D	Hs.180911	ribosomal protein S4, Y-linked	6192	RPS4Y
R27585	134544	Proteasome component C2	10.93	0.89	11	2.04	D	Hs.82159	proteasome (prosome, macropain) subunit, alpha type, 1	5682	PSMA1
AA280846	711552	Carbonyl reductase	11.01	0.77	11	2.44	D	Hs.88778	carbonyl reductase 1	873	CBR1
R91137	195051	ESTs	11.02	0.70	11	2.88	D	Hs.7420	ESTs		
N24004	268727	Human mutY homolog (hMYH) gene, complete cds	11.03	0.78	11	2.21	D	Hs.78489	Human mutY homolog (hMYH) gene, complete cds		
AA406020	742132	INTERFERON-INDUCED 17 KD PROTEIN	11.06	0.83	11	3.76	D	Hs.833	interferon-stimulated protein, 15 kDa	9636	ISG15
T70541	108265	ESTs	11.07	0.76	11	2.08	D	Hs.13740	ESTs		
N50686	280837	ESTs	11.10	0.87	11	2.74	D	Hs.5320	Human Chromosome 16 BAC clone CIT987SK-A-101F10		
AA434305	770837	Human serine/threonine kinase mRNA, partial cds	11.10	0.77	11	2.47	D	Hs.79337	Human serine/threonine kinase mRNA, partial cds		
H93308	241988	ESTs	11.14	0.73	11	2.76	D	Hs.8737	ESTs		
T67069	66555	ESTs	11.18	0.83	11	2.00	D	Hs.13011	ESTs		
H62163	208413	Hepsin	11.26	0.78	11	2.19	D	Hs.823	hepsin (transmembrane protease, serine 1)	3249	HPN
AA733038	399532	Homo sapiens RNA polymerase I subunit hRPA39 mRNA, complete cds	11.27	0.80	11	2.33	D	Hs.5409	RNA polymerase I subunit	9533	RPA40
N67487	291880	Microfibrillar-associated protein 2	11.30	0.76	11	2.15	D	Hs.83551	microfibrillar-associated protein 2	4237	MFAP2
AA454926	814731	Human HBV-X associated (XAP2) mRNA, complete cds	11.32	0.73	11	2.28	D	Hs.75305	aryl hydrocarbon receptor-interacting protein	9049	AIP
R60946 R61067	42313	Prohibitin	11.56	0.78	12	2.27	D	Hs.205821 Hs.75323	ESTs prohibitin		PHB

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Table 1

GenBank ID	Image Clone ID	Clone Name	Cluster location	Max. Correlation	template max	Ratio change	Direction	Unigene	Hs Title	Locus ID	Gene Symbol
AA700604	433350	Sorbitol dehydrogenase	11.77	0.77	12	2.12	D	Hs.878	sorbitol dehydrogenase	6652	SORD
W95082	415145	Hydroxysteroid (11-beta) dehydrogenase 2	12.21	0.73	12	2.32	D	Hs.196726	ESTs		

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Table 2

Template cluster order	Max Corr. Coef.	Fold diff in ratio	IMAGE Clone ID	Accession #	Clone Name	name	Effect of castration
1.012116	0.846729	3.18325	843249	AA486027	Human transcriptional repressor (NAB1) NAB1 mRNA, complete cds	NAB1	INCREASING
1.237519	0.954595	6.13405	298417	N74131	INTESTINAL TREFOIL FACTOR PRECURSOR		INCREASING
1.30759	0.945159	3.10684	815284	AA481543 AA481608	Peptidase D	PEPD PEPD	INCREASING
1.338016	0.94655	4.53497	839101	AA487623	Cardiac gap junction protein	GJA1	INCREASING
1.377788	0.89188	12.0833	153411	R47979	HLA CLASS II HISTOCOMPATIBILITY ANTIGEN, DR ALPHA CHAIN PRECURSOR		INCREASING
2.001098	0.881115	4.45973	243816	N39161	CD36 antigen (collagen type I receptor, thrombospondin receptor)	CD36	INCREASING
2.034041	0.818018	5.15354	489839	AA102107	Glutamyl aminopeptidase (aminopeptidase A)	ENPEP	INCREASING
2.050205	0.877446	3.0482	39920	R53935 R53330	P glycoprotein 3/multiple drug resistance 3	PGY3	INCREASING
2.062313	0.826923	3.48834	343867	W69954	Allograft inflammatory factor 1	AIF1	INCREASING
2.084293	0.940221	3.99105	503083	AA148641	Homo sapiens homeobox protein MEIS2 (MEIS2) mRNA, partial cds	MEIS2	INCREASING
2.128285	0.968211	3.73158	897667	AA496809	SNF2 (sucrose nonfermenting, yeast, homolog)-like 1	SMARCA1	INCREASING
2.133603	0.930549	5.32634	66322	T66799	CD3G antigen, gamma polypeptide (TiT3 complex)	CD3G	INCREASING
2.17829	0.927977	3.77318	195340	R89567 R88884	Group-specific component (vitamin D binding protein)		INCREASING
2.188089	0.981628	6.31585	78294	T50788 T50951	UDP glucuronosyltransferase precursor (UGT2B15)	UGT2B15 UGT2B15	INCREASING
2.898827	0.829225	4.63119	630013	AA219061	DNA repair protein MSH2	MSH2	INCREASING
2.905027	0.889811	3.29801	257162	N30553	Pregnancy-specific beta-1 glycoprotein 4	PSG4	INCREASING
2.915012	0.837463	3.10136	813841	AA447797	Plasminogen activator, tissue type (t-PA)	PLAT	INCREASING
2.917408	0.892144	3.88429	266146	N21576	Human mitochondrial 1,25-dihydroxyvitamin D3 24-hydroxylase mRNA, complete cds		INCREASING
2.942576	0.840711	3.01754	840511	AA486321	Vimentin	VIM	INCREASING
2.945788	0.964988	4.08996	415084	W93369	Cholinergic receptor, nicotinic, alpha polypeptide 7		INCREASING
2.948387	0.933442	5.05904	841695	AA487590	Human BRCA2 region, mRNA sequence CG018		INCREASING
2.971362	0.807993	5.52807	137139	R36006	ESTs		INCREASING
2.971919	0.870528	3.29709	136557	R34604 R34603	ESTs		INCREASING
2.977262	0.978539	3.1673	509731	AA045699	TUMOR-ASSOCIATED ANTIGEN CO-029	TM4SF3	INCREASING

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Table 2

Template cluster order	Max Corr. Coef.	Fold diff in ratio	IMAGE Clone ID	Accession #	Clone Name	name	Effect of castration
2.977769	0.840845	4.56184	840460	AA485865	Interleukin 7 receptor	IL7R	INCREASING
2.978491	0.971108	4.55435	111750	T91100	ESTs		INCREASING
2.978773	0.903384	3.12816	711918	AA282134	H.sapiens mRNA for glutamine cyclotransferase		INCREASING
2.982422	0.871963	4.87652	809998	AA454854	ALPHA-AMYLASE 2B PRECURSOR		INCREASING
2.982555	0.97701	3.30975	784296	AA447079	Mineralocorticoid receptor (aldosterone receptor)	MLR	INCREASING
2.987469	0.865876	3.27205	82734	T73556	Long chain fatty acid acyl-coA ligase	FACL1	INCREASING
2.991463	0.914597	3.88427	322160	W37864	Phosphatase and tensin homolog (mutated in multiple advanced cancers 1	PTEN	INCREASING
2.993909	0.864001	3.33603	177772	H45976	Human cadherin-associated protein-related (cap-r) mRNA, complete cds		INCREASING
3.007232	0.952385	8.30352	223350	H86554	Ceruloplasmin (ferroxidase)	CP	INCREASING
3.009044	0.913093	4.07568	502832	AA128328	Retinoblastoma-binding protein 1{alternative products}	RBBP1	INCREASING
3.011514	0.800143	8.69006	813611	AA447692	Human homeobox gene (clone HHO.c13)		INCREASING
3.012064	0.952765	3.54205	140806	R66310	Peptidylglycine alpha-amidating monooxygenase	PAM	INCREASING
3.01979	0.908171	3.00045	772425	AA405569	Human fibroblast activation protein mRNA, complete cds	FAP	INCREASING
3.032695	0.985948	3.91	246430	N53031	UDP-GLUCURONOSYLTRANSFERASE 2B4 PRECURSOR,		INCREASING
3.043371	0.814047	4.52751	132140	R26163	ESTs		INCREASING
3.047265	0.934034	3.28626	137096	R35979	ESTs		INCREASING
3.056815	0.986131	4.17003	768246	AA424938	Glucose-6-phosphate dehydrogenase	G6PD	INCREASING
3.057154	0.924057	3.77932	897906	AA598652	Sialyltransferase 1 (beta-galactoside alpha-2,6-sialytransferase)	SIAT1	INCREASING
3.060715	0.947831	3.76648	1412481	AA845156	Serine protease inhibitor, Kazal type 1	SPINK1	INCREASING
3.060898	0.971981	3.40674	124071	R02586	ESTs		INCREASING
3.060931	0.970298	3.36991	42070	R60343	5' nucleotidase (CD73)	NT5	INCREASING
3.08443	0.904019	4.63274	362483	AA018780	Spectrin, beta, non-erythrocytic 1	SPTBN1	INCREASING
3.089404	0.824359	3.43277	382195	AA062993	Myosin VIIA (Usher syndrome 1B (autosomal recessive, severe))	MYO7A	INCREASING
3.754562	0.945267	3.28875	130826	R22189	ESTs		INCREASING
3.765124	0.888628	4.09548	811162	AA485748	Fibromodulin	FMOD	INCREASING
3.80855	0.884496	3.65236	796613	AA461456	Collagen, type V, alpha	COL5A2	INCREASING
3.843285	0.816267	3.09501	138936	R62817	ERYTHROCYTE BAND 7 INTEGRAL MEMBRANE PROTEIN	EPB72	INCREASING
3.924886	0.892985	3.42807	811088	AA485665	Human putative EPH-related PTK receptor ligand LERK-8 (Eplg8) mRNA, complete cds	EFNB3	INCREASING
3.971288	0.92074	3.25637	142788	R71093	Human mRNA for collagen binding protein 2, complete cds	CBP2	INCREASING
3.977379	0.944256	5.65444	345935	W77839	Homo sapiens mad protein homolog (hMAD-3) mRNA, complete cds		INCREASING

Table 2

Template cluster order	Max Corr. Coef.	Fold diff in ratio	IMAGE Clone ID	Accession #	Clone Name	name	Effect of castration
4.022205	0.934825	4.06889	773301	AA425217	Cadherin 3 (P-cadherin)	CDH3	INCREASING
4.030049	0.949355	4.83848	781139	AA429895	Human multidrug resistance-associated protein homolog (MRP3) mRNA, partial cds	CMOAT2	INCREASING
4.05323	0.85222	3.68583	810960	AA459401	Homo sapiens serine protease-like protease (nes1) mRNA, complete cds		INCREASING
4.548312	0.865246	5.25408	813823	AA447781	Lumican	LUM	INCREASING
4.550752	0.861665	3.40597	242062	H93837	Apolipoprotein B (including Ag(x) antigen)	APOB	INCREASING
4.633366	0.945709	3.34724	843098	AA488676	Homo sapiens neuronal tissue-enriched acidic protein (NAP-22) mRNA, complete cds		INCREASING
4.638356	0.92024	6.46445	51582	H22826	Human zinc-finger domain-containing protein mRNA, partial cds	LMO7	INCREASING
4.646712	0.856117	5.22275	839991	AA490172	Collagen, type I, alpha-2	COL1A2	INCREASING
4.672971	0.871116	3.13027	840687	AA486365	Mucin 1, transmembrane	MUC1	INCREASING
4.705173	0.920881	32.2623	66731	T64905	Rieger syndrome (solurshin)	PITX2	INCREASING
4.709465	0.809699	5.16377	272327	N32199	Human melanoma antigen recognized by T-cells (MART-1) mRNA	MLANA	INCREASING
4.767416	0.988018	4.66518	121792	T98195 T98194	ESTs		INCREASING
4.838693	0.98735	5.03913	120964	T96123	ESTs		INCREASING
4.906635	0.899493	3.25058	1374571	AA856874	Paired basic amino acid cleaving enzyme (furin, membrane associated receptor protein)	PACE	INCREASING
4.925617	0.808944	3.8469	842836	AA486275	LEUKOCYTE ELASTASE INHIBITOR	ELANH2	INCREASING
4.963333	0.820173	3.36072	360885	AA012867 AA012866	ADP-ribosylation factor 6	ARF6	INCREASING
4.974443	0.953264	5.25845	815774	AA485141	Human Src-like adapter protein mRNA, complete cds		INCREASING
5.089222	0.889221	3.16243	35828	R45640	Diphtheria toxin receptor (heparin-binding epidermal growth factor-like growth factor)	DTR	INCREASING
7.328666	0.827299	4.43384	788256	AA454098	MITOTIC KINESIN-LIKE PROTEIN-1		DECREASING
7.993284	0.857269	4.04703	196303	R92435	ESTs		DECREASING
8.064665	0.995443	4.51081	415089	W93379	H.sapiens nek2 mRNA for protein kinase	NEK2	DECREASING
8.079786	0.895967	4.52618	785707	AA449336	Homo sapiens protein regulating cytokinesis 1 (PRC1) mRNA, complete cds		DECREASING
8.100987	0.859291	3.44151	774446	AA446120	ADRENOMEDULLIN PRECURSOR	ADM	DECREASING

Table 2

Template cluster order	Max Corr. Coef.	Fold diff in ratio	IMAGE Clone ID	Accession #	Clone Name	name	Effect of castration
8.106229	0.969875	4.13575	781047	AA446462	Homo sapiens mitotic checkpoint protein kinase (BUB1) mRNA, complete cds	BUB1	DECREASING
8.121124	0.958895	3.73717	126650	R06900	ESTs		DECREASING
8.179322	0.943909	3.58877	795936	AA460927	H.sapiens mRNA for translin	TSN	DECREASING
8.187876	0.914666	4.01118	131316	R22949	ESTs		DECREASING
8.189608	0.977735	5.06859	700792	AA284072	Human protein phosphatase (KAP1) mRNA, complete cds	CDKN3	DECREASING
8.326993	0.816127	3.72208	324891	W49667	Homo sapiens putative fatty acid desaturase MLD mRNA, complete cds	MLD	DECREASING
8.327641	0.833943	4.80419	811015	AA485377	P55-C-FOS PROTO-ONCOGENE PROTEIN	FOS	DECREASING
8.348061	0.87199	8.07032	207288	H59663	Homo sapiens insulin induced protein 1 (INSIG1) gene, complete cds	INSIG1	DECREASING
8.942278	0.891594	3.23034	204257	H59231	Human metalloprotease/disintegrin/cysteine-rich protein precursor (MDC9) mRNA, complete cds	ADAM9	DECREASING
8.950645	0.970041	5.22441	769921	AA430504	Human cyclin-selective ubiquitin carrier protein mRNA, complete cds	UBCH10	DECREASING
8.958193	0.940671	3.11138	249603	H84871 H85277	Homo sapiens DCHT mRNA, complete cds		DECREASING
8.962524	0.990469	3.46928	207358	H58873	Human (HepG2) glucose transporter gene mRNA, complete cds		DECREASING
8.964165	0.899619	5.15807	435076	AA701455	Human CENP-F kinetochore protein mRNA, complete cds	CENPF	DECREASING
8.981223	0.872284	3.47898	43550	H05914	Human mRNA for lactate dehydrogenase-A (LDH-A, EC 1.1.1.27)	LDHA	DECREASING
8.995491	0.963459	5.83672	416833	W86653	Human 54 kDa progesterone receptor-associated immunophilin FKBP54 mRNA, partial cds	FKBP5	DECREASING
9.017789	0.96145	5.45155	814701	AA481076	Homo sapiens mitotic feedback control protein Madp2 homolog mRNA, complete cds	MAD2L1	DECREASING
9.020971	0.946551	8.47047	898286	AA598974	Cell division cycle 2, G1 to S and G2 to M	CDC2 CDC2	DECREASING
9.032276	0.92293	4.69588	204214	H59204	Human Cdc6-related protein (HsCDC6) mRNA, complete cds	CDC18L	DECREASING
9.036931	0.887974	3.84445	796646	AA461467	Ornithine decarboxylase 1	ODC1	DECREASING
9.037286	0.937001	4.68027	129865	R19158	Homo sapiens mRNA for aurora/IPL1-related kinase, complete cds	STK15	DECREASING
9.042828	0.95762	5.1197	66406	T66935	ESTs		DECREASING
9.043928	0.972186	3.11063	451907	AA706968	H.sapiens mRNA for M-phase phosphoprotein, mpp5		DECREASING
9.077777	0.863953	3.07375	711768	AA280832	Homo sapiens UDP-galactose-4-epimerase (GALE) mRNA, complete cds		DECREASING
9.100562	0.892035	3.68512	416833	W86653	Human 54 kDa progesterone receptor-associated immunophilin FKBP54 mRNA, partial cds	FKBP5	DECREASING
9.656433	0.853914	3.05463	453107	AA700904	Homo sapiens Porc-PI gene similar to yeast CDC45	CDC45L	DECREASING

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Table 2

Template cluster order	Max Corr. Coef.	Fold diff in ratio	IMAGE Clone ID	Accession #	Clone Name	name	Effect of castration
9.663644	0.839201	3.93424	509887	AA056465	Human 54 kDa protein mRNA, complete cds	MLLT7	DECREASING
9.682765	0.845227	6.3141	66728	T64893	Ferrochelatase (protoporphyria)	FECH	DECREASING
9.682844	0.898914	3.51149	789204	AA450205	Human mRNA for translocation protein-1, complete cds	TLOC1	DECREASING
9.742508	0.890147	3.29229	626716	AA191548 AA191245	Human RNA polymerase II elongation factor ELL2, complete cds		DECREASING
9.746844	0.825335	3.39519	47833	H11622	Homo sapiens endothelin-1 (EDN1)	TUBA3	DECREASING
9.767257	0.917964	3.9671	149013	R82300	S-adenosylmethionine decarboxylase 1	AMD1	DECREASING
9.774444	0.872464	7.01222	531319	AA071486	Homo sapiens protein kinase mRNA, complete cds	STK12	DECREASING
9.789542	0.980464	3.0914	789182	AA450265	Proliferating cell nuclear antigen	PCNA	DECREASING
9.791251	0.906944	5.10124	856427	AA630784	Homo sapiens HPV16 E1 protein binding protein mRNA, complete cds		DECREASING
9.795647	0.941355	6.1827	725454	AA397813	CDC28 protein kinase 2	CKS2	DECREASING
9.819214	0.85287	3.01659	49352	H15446	Annexin VII (synexin)	ANX7	DECREASING
9.82745	0.865398	4.0672	293727	N69694	ESTs		DECREASING
9.82892	0.899518	3.3255	273546	N33274	MULTIFUNCTIONAL PROTEIN ADE2		DECREASING
9.837101	0.82697	3.27689	53316	R15814	Human malate dehydrogenase (MDHA) mRNA, complete cds	MDH1	DECREASING
9.838391	0.90832	3.43685	42059	R60317	Human dihydrolipoamide dehydrogenase mRNA, complete cds	DLD	DECREASING
9.867489	0.934121	3.48858	855487	AA664155	Human putative 32kDa heart protein PHP32 mRNA, complete cds	ASAH	DECREASING
9.870099	0.819685	5.05212	281003	N50880	T cell receptor gamma chain	TCRG	DECREASING
9.884679	0.925499	3.43577	898062	AA598776	Human p55CDC mRNA, complete cds	CDC20	DECREASING
9.886957	0.842022	3.17901	24145	R37953	Homo sapiens adenylyl cyclase-associated protein (CAP) mRNA, complete cds	CAP	DECREASING
9.942197	0.894898	3.67595	134719	R28287 R28071	ESTs		DECREASING
9.945983	0.86581	3.20405	684655	AA251770	26S PROTEASE REGULATORY SUBUNIT 7	PSMC2	DECREASING
9.954306	0.902556	3.1436	29063	R40970 R14475	ESTs		DECREASING
9.969233	0.805497	3.0064	45233	H07880	Human chaperonin protein (Tcp20) gene complete cds	CCT6	DECREASING
9.971725	0.817539	4.00407	814117	AA465386	Human Gu protein mRNA, partial cds		DECREASING
10.012323	0.942758	3.67632	283315	N54768	Phosphoglycerate mutase 2 (muscle)	PGAM2	DECREASING
10.062454	0.866873	3.02895	785778	AA449693 AA448967	Homo sapiens spleen mitotic checkpoint BUB3 (BUB3) mRNA, complete cds	BUB3 BUB3	DECREASING

Table 2

Template cluster order	Max Corr. Coef.	Fold diff in ratio	IMAGE Clone ID	Accession #	Clone Name	name	Effect of castration
10.642527	0.938567	7.09894	840567	AA487893	TUMOR-ASSOCIATED ANTIGEN L6	TM4SF1	DECREASING
10.720656	0.948241	3.096	767817	AA418689	DNA-DIRECTED RNA POLYMERASE II 14.4 KD POLYPEPTIDE		DECREASING
11.056153	0.828701	3.7635	742132	AA406020	INTERFERON-INDUCED 17 KD PROTEIN	ISG15	DECREASING

Hierarchical cluster position	Ratio Fold Change	Clone Index	clone name	unigene name	accession
1	2.068256	531957	Human mRNA for ubiquitin-conjugating enzyme, complete cds	UBE2G1	AA113903
2	3.073747	711768	Homo sapiens UDP-galactose-4-epimerase (GALE) mRNA, complete cds		AA280832
3	2.645818	971367	40S RIBOSOMAL PROTEIN S8	RPS8	AA683050
4	2.539682	129146	Homo sapiens mRNA for COX7RP, complete cds	COX7RP	R10947
5	2.505608	825677	Homo sapiens arsenite translocating ATPase (ASNA1) mRNA, complete cds	ASNA1	AA504809
7	2.628698	1E+06	40S RIBOSOMAL PROTEIN S15A	RPS15A	AA872341
8	3.179007	24145	Homo sapiens adenylyl cyclase-associated protein (CAP) mRNA, complete cds	CAP	R37953
9	3.20405	684655	26S PROTEASE REGULATORY SUBUNIT	PSMC2	AA251770
10	3.016585	49352	Annexin VII (synexin)	ANX7	H15446
11	5.995891	162775	P68 PROTEIN	DDX5	H27646
12	2.2082	108377	Tubulin, gamma polypeptide	TUBG	T77732
13	3.013561	812965	V-myc avian myelocytomatosis viral oncogene homolog	MYC	AA464600
14	2.839618	244154	ESTs		N52450
15	1.772225	781097	Human protein tyrosine kinase t-Ror1 (Ror1) mRNA, complete cds	NTRKR1	AA430035
16	3.198344	324342	ESTs		W47576 W47597
17	2.480446	43231	Human mRNA for 26S proteasome subunit p97	PSMD2	H05893
20	2.600338	545403	Homo sapiens thioredoxin-like protein mRNA, complete cds	TXNL	AA079059
23	2.76642	824568	Prostate specific antigen	KLK3	AA490981
24	2.235154	810057	DNA-BINDING PROTEIN A	CSDA	AA465019
25	3.313153	897774	Human APRT gene for adenine phosphoribosyltransferase	APRT	AA598510
26	3.006397	45233	Human chaperonin protein (Tcp20) gene complete cds	CCT6	H07880
27	2.700649	840702	Human selenium donor protein (selD) mRNA, complete cds		AA488081
28	2.306756	376785	Homo sapiens mRNA for putative progesterone binding protein		AA047567
30	4.00407	814117	Human Gu protein mRNA, partial cds		AA465386
31	2.0077	878130	H.sapiens mRNA for SMT3B protein		AA775415
32	4.62198	785293	H.sapiens mRNA for rat HREV107-like protein		AA476438
34	2.433838	428223	Homo sapiens EB1 mRNA, complete cds	HMG1	AA001819
36	2.973092	487348	Homo sapiens dynamin-like protein mRNA, complete cds	DYMPLE	AA045529
37	2.921066	951142	FLAP ENDONUCLEASE-1	FEN1	AA620553
38	2.819821	950690	Cyclin A	CCNA2	AA608568
39	3.469276	207358	Human (HepG2) glucose transporter gene mRNA, complete cds		H58873
40	2.660381	884867	Eukaryotic translation initiation factor 5 (eIF5)	EIF5	AA669443
42	4.510814	415089	H.sapiens nek2 mRNA for protein kinase	NEK2	W93379
44	4.52618	785707	Homo sapiens protein regulating cytokinesis 1 (PRC1) mRNA, complete cds		AA449336

Hierarchical cluster position	Ratio Fold Change	Clone Index	clone name	unigene name	accession
46	2.240431	295483	ESTs		N70382
47	1.963496	868308	40S RIBOSOMAL PROTEIN S23	RPS23	W05026
48	2.278726	756401	H.sapiens mRNA for ras-related GTP-binding protein	RHEB2	AA634008 AA482117 AA482441
49	2.639337	813675	Human D9 splice variant A mRNA, complete cds		
50	2.665981	44537	Unknown EST	SNRPB	AA453750 H06853
51	2.223912	795738	Human G protein gamma-10 subunit mRNA, complete cds	GNG10	AA460286
52	2.450932	755239	H.sapiens mRNA for D1075-like gene	METTL1	AA422058
54	2.287996	234237	H.sapiens mRNA for Pirin, isolate 1	NME2 PIR	AA422139 H69335
55	2.533715	772304	Adenine nucleotide translocator 2 (fibroblast)	ANT2	AA404486
56	5.068587	700792	Human protein phosphatase (KAP1) mRNA, complete cds	CDKN3	AA284072
57	2.402217	853368	Thymidylate synthase		AA663310
58	2.363695	878676	ATP-DEPENDENT DNA HELICASE II, 86 KD SUBUNIT	XRCC5	AA775355
59	3.967096	149013	S-adenosylmethionine decarboxylase 1	AMD1	R82300
60	2.478747	757489	Homo sapiens (clone ch13lambda7) alpha-tubulin mRNA, complete cds	TUBA2	AA426374 AA436990
61	3.230339	204257	Human metalloprotease/disintegrin/cysteine-rich protein precursor (MDC9) mRNA, complete cds	ADAM9	H59231
62	3.081688	897567	L-LACTATE DEHYDROGENASE M CHAIN	LDHA	AA497029
63	3.47898	43550	Human mRNA for lactate dehydrogenase-A (LDH-A, EC 1.1.1.27)	LDHA	H05914
64	2.54226	725188	MALATE DEHYDROGENASE, CYTOPLASMIC	MDH1	AA403295
65	3.844452	796646	Ornithine decarboxylase 1	ODC1	AA461467
66	3.054629	453107	Homo sapiens Porc-PI gene similar to yeast CDC45	CDC45L	AA700904
68	2.688569	753457	NADH-UBIQUINONE OXIDOREDUCTASE 75 KD SUBUNIT PRECURSOR	NDUFS1	AA406536
69	3.276888	53316	Human malate dehydrogenase (MDHA) mRNA, complete cds	MDH1	R15814
70	7.012224	531319	Homo sapiens protein kinase mRNA, complete cds	STK12	AA071486
71	3.436845	42059	Human dihydrolipoamide dehydrogenase mRNA, complete cds	DLD	R60317
72	5.101236	856427	Homo sapiens HPV16 E1 protein binding protein mRNA, complete cds		AA630784
73	3.395189	47833	Homo sapiens endothelin-1 (EDN1)	TUBA3	H11622
75	4.333755	809588	Homo sapiens human gamma-glutamyl hydrolase (hGH) mRNA, complete cds	GGH	AA456621
76	3.325499	273546	MULTIFUNCTIONAL PROTEIN ADE2		N33274
77	2.378417	950482	Small nuclear ribonucleoprotein polypeptides B and B1	SNRPB	AA599116

Hierarchical cluster position	Ratio Fold Change	Clone Index	clone name	unigene name	accession
78	2.674778	814615	NAD-DEPENDENT METHYLENETETRAHYDROFOLATE DEHYDROGENASE		AA480995
80	3.511485	789204	Human mRNA for translocation protein-1, complete cds	TLOC1	AA450205
81	4.695879	204214	Human Cdc6-related protein (HsCDC6) mRNA, complete cds	CDC18L	H59204
82	5.836716	416833	Human 54 kDa progesterone receptor-associated immunophilin FKBP54 mRNA, partial cds	FKBP5	W86653
82	3.685118	416833	Human 54 kDa progesterone receptor-associated immunophilin FKBP54 mRNA, partial cds	FKBP5	W86653
83	3.091404	789182	Proliferating cell nuclear antigen	PCNA	AA450265
84	3.110629	451907	H.sapiens mRNA for M-phase phosphoprotein, mpp5		AA706968
85	2.600775	44975	Human homolog of yeast IPP isomerase	IDI1	H08820
86	5.451547	814701	Homo sapiens mitotic feedback control protein Madp2 homolog mRNA, complete cds	MAD2L1	AA481076
87	5.224413	769921	Human cyclin-selective ubiquitin carrier protein mRNA, complete cds	UBCH10	AA430504
89	6.182703	725454	CDC28 protein kinase 2	CKS2	AA397813
90	8.470467	898286	Cell division cycle 2, G1 to S and G2 to M	CDC2 CDC2	AA598974
93	1.943555	271006	Dihydrolipoamide S-acetyltransferase (E2 component of pyruvate dehydrogenase complex)	DLAT	N42953
95	2.512932	295986	H.sapiens mRNA for phenylalkylamine binding protein		N67038
96	4.011178	131316	ESTs		R22949
97	3.588767	795936	H.sapiens mRNA for translin	TSN	AA460927
98	8.070317	207288	Homo sapiens insulin induced protein 1 (INSIG1) gene, complete cds	INSIG1	H59663
99	2.330458	745138	Human alpha-tubulin isotype H2-alpha gene, last exon	TUBA2	AA626698
100	2.71733	200402	ESTs		R96998
101	2.130216	813648	Dihydrolipoamide dehydrogenase (E3 component of pyruvate dehydrogenase complex, 2-oxo-glutarate complex, branched chain keto acid dehydrogenase complex)	DLD	AA447748
103	2.035546	840364	S-adenosylhomocysteine hydrolase	AHCY	AA485626
104	2.142688	856489	RIBONUCLEOSIDE-DIPHOSPHATE REDUCTASE M1 CHAIN	RRM1	AA633549
105	3.435773	898062	Human p55CDC mRNA, complete cds	CDC20	AA598776
107	2.210029	626206	DNA polymerase gamma		AA188761
108	2.387246	813280	Adenylosuccinate lyase	ADSL	AA455931
109	3.095995	767817	DNA-DIRECTED RNA POLYMERASE II 14.4 KD POLYPEPTIDE		AA418689
111	2.441951	433666	H.sapiens mRNA for testican	SPOCK	AA699317
112	7.098935	840567	TUMOR-ASSOCIATED ANTIGEN L6	TM4SF1	AA487893

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113	2.711239	739511	Human kinase Myt1 (Myt1) mRNA, complete cds	PKMYT1	AA479030
114	2.780629	810552	Human B-cell receptor associated protein (hBAP) mRNA, partial cds		AA464567
115	3.676315	283315	Phosphoglycerate mutase 2 (muscle)	PGAM2	N54768
116	1.924043	40017	CYTOCHROME C	CYC1	R53311
116	2.996875	40017	Cytochrome c-1	CYC1	R53311
117	2.71995	46171	Human mRNA for eukaryotic initiation factor 4AI		H09590
118	3.488577	855487	Human putative 32kDa heart protein PHP32 mRNA, complete cds	ASAH	AA664155
120	2.556136	1160558	6-PYRUVOYL TETRAHYDROBIOPTERIN SYNTHASE	PTS	AA877347
121	3.028945	785778	Homo sapiens spleen mitotic checkpoint BUB3 (BUB3) mRNA, complete cds	BUB3 BU B3	AA449693 AA448967
122	2.366766	246120	Human mRNA for suppressor for yeast mutant, complete cds	HRMT1L2	N55480
123	2.702014	325641	Pregnancy specific beta-1 glycoprotein 5	PSG5	W51985 W52627
125	1.975916	586650	Human placental equilibrative nucleoside transporter 1 (hENT1) mRNA, complete	ENT1	AA129135
126	2.175796	324618	MITOCHONDRIAL ELONGATION FACTOR TS PRECURSOR	TSFM	W47014
127	1.92211	813712	ATP synthase, H+ transporting, mitochondrial F0 complex, subunit b, isoform 1	ATP5F1	AA453765
128	2.481154	797016	Succinate dehydrogenase 1, iron sulphur (Ip) subunit	SDHB	AA463510
140	2.114314	308682	Homo sapiens mRNA for putative ABC transporter, partial		N95462
141	2.266048	80946	Ribonuclease L (2',5'-oligoisoadenylate synthetase-dependent) inhibitor	RNASELI	T70056
142	3.19801	144762	ESTs		R77213
143	2.113186	322914	Acid phosphatase 1, soluble	ACP1	W45148
144	2.582853	611150	Human mitochondrial ATP synthase subunit 9, P3 gene copy, mRNA, nuclear gene encoding mitochondrial protein, complete cds	ATP5G3	AA173109 AA173369
145	1.715618	248531	Human guanosine 5'-monophosphate synthase mRNA, complete cds	GMPS	N59764
147	2.369768	744047	Human pLK mRNA, complete cds	PLK	AA629262
149	2.239031	362926	Protein kinase, cAMP-dependent, catalytic, beta	PRKACB	AA018980
151	3.734653	810711	Cytochrome B561	CYB561	AA457700 AA480809
153	3.292291	626716	Human RNA polymerase II elongation factor ELL2, complete cds		AA191548 AA191245
157	2.858839	783697	Homo sapiens E1B 19K/Bcl-2-binding protein Nip3 mRNA, nuclear gene encoding mitochondrial protein, complete	BNIP3	AA446839
160	1.882036	700721	CDC46 HOMOLOG	MCM5	AA285155
165	1.795108	32898	Human mRNA for mitochondrial short-chain enoyl-CoA hydratase, complet	ECHS1	R43558

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166	2.021158	321661	Protein phosphatase 2A, regulatory subunit B' alpha-1		W35378
167	2.788854	46182	CTP synthetase	CTPS	H09614
168	2.064348	826211	Programmed cell death 2	PDCD2	AA521466
169	2.088606	869538	Human NADH:ubiquinone oxidoreductase MLRQ subunit mRNA, complete cds	NDUFA4	AA680322
171	2.093819	45544	SM22-ALPHA HOMOLOG	TAGLN2	H08564
172	2.382227	309288	Replication factor C, 37-kD subunit	RFC4	N93924
173	1.55836	877835	Human ribosomal protein L35 mRNA, complete cds		AA625634
174	1.688785	83363	Human mRNA for PIMT isozyme I, complete cds	PCMT1	T68453
177	3.698942	26578	Homo sapiens pescadillo mRNA, complete cds		R37665
179	2.324977	509495	PROTEASOME IOTA CHAIN	PSMA6	AA047319
180	1.880246	1033708	GAMMA CRYSTALLIN A		AA780079
181	1.99955	47681	Human putative splice factor transformer2-beta mRNA, complete cds		H11720 H11792
182	2.167164	950473	Homo sapiens BAF57 (BAF57) gene, complete cds	SMARCE 1	AA599120
183	2.676983	626531	Homo sapiens RRM RNA binding protein Gry-rbp (GRY-RBP) mRNA, complete cds	NSAP1	AA186327
184	2.417391	845363	NUCLEOSIDE DIPHOSPHATE KINASE A	NME1	AA644092
187	2.152056	234562	Homo sapiens splicing factor Sip1 mRNA, complete cds	SRRP129	H78241
188	1.962195	280507	Hypoxanthine phosphoribosyltransferase 1 (Lesch-Nyhan syndrome)	HPRT1	N47312
189	2.510673	838802	Procollagen-proline, 2-oxoglutarate 4-dioxygenase (proline 4-hydroxylase), alpha polypeptide	P4HA1	AA464908
190	2.737453	34778	Vascular endothelial growth factor	VEGF	R45059 R19956
192	2.647888	785616	Signal sequence receptor, alpha	SSR1	AA450360
193	2.596588	320509	Homo sapiens mRNA for cytochrome b5, partial cds		W04674 W31775
194	2.894434	51448	Activating transcription factor 3	ATF3	H21042
197	2.493428	725284	Phosphorylase kinase, gamma 2 (testis)		AA291732
198	2.08295	612274	TUBULIN ALPHA-4 CHAIN	TUBA1	AA180742

No.	Clone ID	Cal. Ratio	Recur. Intensity	Primary Intensity	Description
1	179276	9.612	16375.6	1513.5	Human breast carcinoma fatty acid synthase mRNA, complete cds
2	770212	9.354	3499.5	332.4	CARTILAGE GLYCOPROTEIN-39 PRECURSOR
3	135221	8.871	46969.3	4703.7	S-100P PROTEIN
4	140574	7.388	19367.4	2328.9	Human CX3C chemokine precursor, mRNA, alternatively spliced, complete cds
5	297061	5.173	3159	542.5	Homo sapiens mRNA for dihydropyrimidinase, complete cds
6	129342	4.887	19608.4	3564.8	ESTs
7	180864	4.067	3943.5	861.5	Human telencephalin precursor mRNA, complete cds
8	839094	3.956	3130	702.9	Human beta-A3/A1 crystallin (CYRBA3/A1) mRNA, partial cds
9	295483	3.903	23246.8	5291.6	ESTs
10	53039	3.829	4303.4	998.5	Homo sapiens chondroitin-6-sulfotransferase mRNA, complete cds
11	78294	3.612	11393.9	2802.2	UDP glucuronosyltransferase precursor (UGT2B15)
12	361974	3.611	8046	1979.7	Pleiotrophin (heparin binding growth factor 8, neurite growth-promoting factor 1)
13	194307	3.532	3434.1	863.9	ESTs
14	1460110	3.441	6511	1681.2	Proteasome (prosome, macropain) subunit, beta type, 5
15	212640	3.424	2895.7	751.4	H.sapiens partial C1 mRNA
16	453689	3.274	14395.4	3905.8	Homo sapiens mRNA for rab geranylgeranyl transferase, alpha-subunit
17	340734	3.227	4565.1	1256.9	MAJOR HISTOCOMPATIBILITY COMPLEX ENHANCER-BINDING PROTEIN MAD3
18	742132	3.159	17358.3	4882.1	INTERFERON-INDUCED 17 KD PROTEIN
19	41650	3.156	4626.4	1302.3	Hepatocyte growth factor (hepapoietin A; scatter factor)
20	812251	2.967	25336.8	7587.3	H.sapiens mRNA for MAP kinase activated protein kinase
21	795730	2.958	4436.9	1332.5	Homo sapiens mRNA for Efs1, complete cds
22	1472689	2.946	2891.3	872	Apolipoprotein CI
23	768562	2.866	27406.3	8494.7	Human hkf-1 mRNA, complete cds
24	724615	2.848	14876.5	4640.8	Chromosome condensation 1
25	322723	2.836	3054.3	956.8	ESTs
26	135630	2.82	24116.9	7598.1	Human CDP-diacylglycerol synthase (CDS) mRNA, complete cds
27	839101	2.769	22037	7071.1	Cardiac gap junction protein
28	50794	2.705	4548.9	1494	Zinc finger protein 133 (clone pHZ-13)
29	712049	2.659	3577.3	1195.3	Human MDA-7 (mda-7) mRNA, complete cds
30	366067	2.636	3097.1	1043.7	Cerebellar degeneration-related protein (62kD)
31	256907	2.621	3579	1213.3	Glutathione S-transferase A3
32	122955	2.612	6652.3	2262.5	ESTs
33	429466	2.594	3672.6	1258	Homo sapiens mRNA for synaptogyrin 1a
34	841641	2.58	7321.5	2521.4	Cyclin D1 (PRAD1; parathyroid adenomatosis 1)
35	503097	2.572	8971.3	3098.8	Phosphoribosyl pyrophosphate synthetase 2
36	754600	2.545	4929.8	1721.1	Nuclear factor I/X (CCAAT-binding transcription factor)

No.	Clone ID	Cal. Ratio	Recur. Intensity	Primary Intensity	Description
37	756405	2.524	13892.6	4889.5	Inhibitor of DNA binding 3, dominant negative helix-loop-helix protein
38	139250	2.521	3846.5	1355.6	ESTs
39	1475595	2.508	4175	1478.8	Alkaline phosphatase, liver/bone/kidney
40	66317	2.468	10365.6	3731.1	HISTONE H1D
41	139331	2.458	4342.2	1569.6	ESTs
42	810873	2.456	3001.4	1085.7	Solute carrier family 9 (sodium/hydrogen exchanger), isoform 1 (antiporter, Na+/H+, amiloride sens
43	796475	2.448	2929.5	1062.9	Homo sapiens skeletal muscle LIM-protein FHL3 mRNA, complete cds
44	810711	2.432	15015.7	5486.1	Cytochrome B561
45	755578	2.412	9607.2	3538.7	INTEGRAL MEMBRANE PROTEIN E16
46	150702	2.372	2994.7	1121.6	Homeo box B5 (2.1 protein)
47	293104	2.326	5386.5	2057.7	Homo sapiens peroxisomal phytanoyl-CoA alpha-hydroxylase (PAHX) mRNA, complete cds
48	143995	2.31	3737.3	1437.2	ESTs
49	487172	2.3	3648.5	1409.5	N-ACETYLLACTOSAMINE SYNTHASE
50	270136	2.286	3109.7	1208.5	Homo sapiens mRNA for leukemia associated gene 2
51	504226	2.282	4037.8	1571.7	CD53 antigen
52	292392	2.251	5437.2	2146	ESTs
53	687820	2.246	2886.2	1141.8	ATPase, Cu++ transporting, alpha polypeptide (Menkes syndrome)
54	300137	2.245	3066.1	1213.4	Homo sapiens mRNA for AMP-activated protein kinase beta 2 subunit
55	1323448	2.23	3092.1	1231.9	Human cysteine-rich heart protein (hCRHP) mRNA, complete cds
56	1049033	2.225	4576.6	1827.7	Homo sapiens mRNA for calmegin, complete cds
57	510381	2.224	5238.7	2092.7	Human DNA-binding protein CPBP (CPBP) mRNA, partial cds
58	296032	2.213	2978.5	1195.9	Type 3 iodothyronine deiodinase
59	143287	2.209	3246.7	1305.6	Pregnancy-specific beta-1 glycoprotein 13
60	278242	2.207	3221.6	1296.6	Homo sapiens pyruvate dehydrogenase kinase isoenzyme 3 (PDK3) mRNA, complete cds
61	727147	2.161	2983	1226.4	SQUAMOUS CELL CARCINOMA ANTIGEN 1
62	773567	2.15	4990.7	2062.1	Protein tyrosine phosphatase, non-receptor type 2
63	143756	2.149	6502.1	2687.6	ESTs
64	236034	2.14	3534.1	1467.2	Uncoupling protein 2 (mitochondrial, proton carrier)
65	841470	2.131	10862.9	4529.2	CATHEPSIN H PRECURSOR
66	768260	2.123	10965.2	4589.3	RETINOBLASTOMA BINDING PROTEIN 3
67	308041	2.116	5624.2	2360.9	Glycoprotein Ib (platelet), beta polypeptide
68	243360	2.105	3422.1	1444.6	ESTs
69	504774	2.092	2952.2	1253.6	GAMMA-GLUTAMYLTRANSPEPTIDASE 5 PRECURSOR
70	280371	2.076	3017.7	1291.2	5-hydroxytryptamine (serotonin) receptor 2C
71	345232	2.072	4821.6	2067	Lymphotoxin alpha (formerly tumor necrosis factor beta)

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72	174627	2.065	2858.9	1230.1	SECRETOGRANIN II PRECURSOR
73	703581	2.064	2971.6	1279.1	Hematopoetic proteoglycan core protein
74	223350	2.062	6154.5	2651.7	Ceruloplasmin (ferroxidase)
75	782339	2.051	3228.5	1398.8	Homo sapiens mRNA for AMP-activated protein kinase beta-1
76	70827	2.044	10586.3	4600.7	Amiloride binding protein 1 (amine oxidase (copper-containing))
77	796646	2.038	36042.8	15710	Ornithine decarboxylase 1
78	415817	2.034	4452.1	1944.8	Cytochrome P450, subfamily IVA, polypeptide 11
79	235040	2.029	5949.2	2605.2	ESTs
80	1456424	2.015	3683.9	1624.1	Human mRNA for alanine aminotransferase
81	195051	2.011	18861.8	8332.7	ESTs
82	44505	2.009	4409.1	1949.9	Human NAD+-dependent succinate-semialdehyde dehydrogenase (SSADH) mRNA, 3' end
83	211813	1.979	5745.4	2579.5	ESTs
84	754509	1.977	5203.7	2338.2	Met proto-oncogene (hepatocyte growth factor receptor)
85	66731	1.977	5147	2313.3	Rieger syndrome (solurshin)
86	245920	1.965	4117.5	1861.5	Glycogen synthase [human, liver, mRNA, 2912 nt]
87	669435	1.963	3596.8	1628	Human C-1 mRNA, complete cds
88	243343	1.955	3370.3	1531.7	Homo sapiens chaperonin containing t-complex polypeptide 1, beta subunit (Cctb) mRNA, complete cds
89	343744	1.943	11043.4	5049.6	Homo sapiens adenosine triphosphatase mRNA, complete cds
90	366541	1.933	3417.2	1570.1	Chymotrypsin-like
91	502682	1.932	16161.3	7432.1	Human enigma gene, complete cds
92	435330	1.92	5590.1	2587	Syntrophin, alpha (dystrophin-associated protein A1, 59kD, acidic component)
93	1470048	1.917	3098.9	1436.2	Human retinoic acid induced RIG-E precursor (E) mRNA, complete cds
94	724831	1.916	12664	5871.4	B cell lymphoma protein 7B
95	814353	1.916	4458.1	2067.7	ATL-derived PMA-responsive (APR) peptide
96	809910	1.894	7020.9	3293.1	INTERFERON-INDUCIBLE PROTEIN 1-8U
97	265874	1.884	8309.3	3917.7	CCAAT BOX-BINDING TRANSCRIPTION FACTOR 1
98	431296	1.878	4298	2032.8	Protein phosphatase 3 (formerly 2B), catalytic subunit, alpha isoform (calcineurin A alpha){altern
99	742862	1.87	4533.3	2153.5	Homo sapiens imprinted multi-membrane spanning polyspecific transporter-related protein (IMPT1) mR
100	784910	1.867	8274.7	3937.1	Human fetus brain mRNA for membrane glycoprotein M6, complete cds
101	268876	1.851	4638.6	2226.7	Homo sapiens survival of motor neuron protein interacting protein 1 (SIP1) mRNA, complete cds
102	713886	1.839	5032.9	2430.8	Human adult heart mRNA for neutral calponin, complete cds
103	240151	1.83	3906.4	1896.5	Inhibitor of DNA binding 2, dominant negative helix-loop-helix protein
104	292222	1.822	4259.1	2076.5	Dihydrolipoamide S-acetyltransferase (E2 component of pyruvate dehydrogenase complex)
105	281978	1.82	3770.9	1841	Homo sapiens Rac3 (RAC3) mRNA, complete cds

No.	Clone ID	Cal. Ratio	Recur. Intensity	Primary Intensity	Description
106	434833	1.817	2838.9	1388.4	PROTEIN 4.1
107	628336	1.812	12578.9	6166.8	Myosin, light polypeptide 1, alkali; skeletal, fast
108	346696	1.811	5223.8	2563.1	Human transcription factor RTEF-1 (RTEF1) mRNA, complete cds
109	141316	1.808	3158.6	1552	ESTs
110	143919	1.803	2803	1381.5	ESTs
111	853809	1.801	4476.4	2208.1	Homo sapiens mRNA for RGS5, complete cds
112	51582	0.554	4764.1	7639.7	Human zinc-finger domain-containing protein mRNA, partial cds
113	143306	0.553	15321.3	24624.5	LYMPHOCYTE-SPECIFIC PROTEIN LSP1
114	246304	0.553	7493.4	12048.6	Human mRNA for tob family, complete cds
115	123980	0.552	4907.9	7903.5	Homo sapiens mRNA for HYA22, complete cds
116	810117	0.549	10136.8	16415.4	Annexin XI (56kD autoantigen)
117	245235	0.546	4105.3	6685.5	ESTs
118	241985	0.543	3190.3	5219.3	Homo sapiens IPW mRNA sequence
119	815555	0.543	2673.3	4374.6	Diacylglycerol kinase, alpha (80kD)
120	128126	0.54	4585.3	7547.8	Decay accelerating factor for complement (CD55, Cromer blood group system)
121	195132	0.539	3233	5329.6	ESTs
122	470393	0.536	2150.3	3561.4	Matrix metalloproteinase 7 (matrilysin, uterine)
123	810703	0.529	21355.6	35863.1	Human high density lipoprotein binding protein (HBP) mRNA, complete cds
124	511521	0.526	15060.2	25426.7	Calnexin
125	133130	0.523	1933.3	3282.4	ESTs
126	469969	0.522	11975.6	20384	Integrin, alpha V (vitronectin receptor, alpha polypeptide, antigen CD51)
127	233274	0.518	6950.7	11925.5	ESTs
128	753184	0.518	5394.2	9258.3	SRY (sex-determining region Y)-box 9 (campomelic dysplasia, autosomal sex-reversal)
129	196303	0.512	6504.3	11295.4	ESTs
130	725877	0.511	17254.5	30001.8	Homo sapiens creatine transporter mRNA, complete cds
131	504791	0.508	21272.1	37217.5	Homo sapiens glutathione transferase (GSTA4) mRNA, complete cds
132	725877	0.507	8767.1	15376	Clusterin (complement lysis inhibitor; testosterone-repressed prostate message 2; apolipoprotein J)
133	144747	0.505	20340.4	35757.7	ESTs
134	234469	0.501	3372.6	5975.1	ESTs
135	1309620	0.501	1929.3	3421.5	Homo sapiens Tax interaction protein 33 mRNA, partial cds
136	897768	0.49	3657.3	6625.4	Alpha-1 type VII collagen
137	784772	0.489	3730.6	6780.1	GRAVIN
138	243291	0.487	4138	7549.1	ESTs
139	756708	0.481	1722	3179.6	Homo sapiens intermediate conductance calcium-activated potassium channel (hKCa4) mRNA, complete c
140	34778	0.477	8673.5	16146.5	Vascular endothelial growth factor
141	417424	0.473	4977.9	9344.7	Human DNA-binding protein ABP/ZF mRNA, complete cds

No.	Clone ID	Cal. Ratio	Recur. Intensity	Primary Intensity	Description
142	298417	0.473	2870.1	5393	INTESTINAL TREFOIL FACTOR PRECURSOR
143	324891	0.463	23336.5	44772.6	Homo sapiens putative fatty acid desaturase MLD mRNA, complete cds
144	160485	0.463	1484.5	2848.3	ESTs
145	200604	0.459	2840.9	5493.1	ESTs
146	743230	0.459	7584.5	14695.1	T3 receptor-associating cofactor-1 [human, fetal liver, mRNA, 2930 nt]
147	130280	0.458	3691.6	7157.8	Human cAMP responsive element binding protein beta subunit (CREBPA) mRNA, complete cds
148	811162	0.456	1554.3	3025.7	Fibromodulin
149	203514	0.451	2398.4	4720	ESTs
150	950682	0.45	1629.4	3217.4	Phosphofructokinase, platelet
151	244955	0.448	4631.4	9181	ESTs
152	841507	0.442	4261.5	8568.6	PULMONARY SURFACTANT-ASSOCIATED PROTEIN A PRECURSOR
153	120375	0.441	2738	5511.1	ESTs
154	187147	0.44	2031.8	4101	Human ras inhibitor mRNA, 3' end
155	28469	0.439	12401.6	25095.9	Human succinyl CoA:3-oxoacid CoA transferase precursor (OXCT) mRNA, complete cds
156	292749	0.439	4604.9	9329.4	ESTs
157	884783	0.437	2668.5	5421	Human PTPL1-associated RhoGAP mRNA, complete cds
158	753587	0.433	3242.7	6647.3	Human butyrophilin protein (BT3.3) mRNA, partial cds
159	564621	0.432	2325.9	4786.5	H.sapiens mRNA for protease inhibitor 12 (PI12, neuroserpin)
160	39093	0.429	4070.6	8426.8	Human eIF-2-associated p67 homolog mRNA, complete cds
161	866702	0.417	1383	2944.9	Human protein tyrosine phosphatase 1E (PTP1E) mRNA, complete cds
162	246722	0.411	2591.9	5602.5	Homo sapiens CAGH3 mRNA, complete cds
163	23185	0.41	4533.1	9832.5	Hexabrachion (tenascin C, cytotactin)
164	377048	0.405	3374.1	7395.6	Homo sapiens incomplete cDNA for a mutated allele of a myosin class I, myh-1c
165	825740	0.404	12627.7	27781.2	Human terminal transferase mRNA, complete cds
166	810512	0.399	3213.9	7147.3	Thrombospondin 1
167	783696	0.385	17731.1	40947.4	Ornithine aminotransferase (gyrate atrophy)
168	26184	0.377	8637.2	20366.5	Human mRNA for platelet-type phosphofructokinase, complete cds
169	502690	0.376	1821.2	4308.3	Ribophorin I
170	713145	0.375	10364.8	24538.6	CD44 antigen (cell adhesion molecule)
171	193087	0.374	13115.4	31189.5	Human hepatocyte growth factor-like protein homolog (D1F15S1A) gene, complete cds
172	308231	0.366	3954	9590.9	ESTs
173	42627	0.362	3476.8	8527.4	Homo sapiens Coch-5B2 mRNA, complete cds
174	757222	0.359	21821.8	53929.5	Human clone HSH1 HMG CoA synthase mRNA, partial cds
175	197323	0.351	1477.2	3737.3	ESTs
177	204735	0.345	2220.5	5725.9	ESTs

No.	Clone ID	Cal. Ratio	Recur. Intensity	Primary Intensity	Description
178	194906	0.34	3263.3	8518.9	ESTs
179	292312	0.34	2212.4	5777.5	ESTs
180	233299	0.334	4134.7	10998.2	ESTs
181	200814	0.331	7705	20660.7	Membrane metallo-endopeptidase (neutral endopeptidase, enkephalinase, CALLA, CD10)
182	144797	0.33	1394.1	3749	ESTs
183	167076	0.33	3076	8292.9	ESTs
184	42864	0.328	3816.2	10341.4	Collagen, type IV, alpha 5 (Alport syndrome)
185	134495	0.326	6353.2	17303.3	ESTs
186	239835	0.323	3322.4	9151.9	ESTs
187	343987	0.3	1809.5	5353.9	Dipeptidylpeptidase IV (CD26, adenosine deaminase complexing protein 2)
188	292391	0.273	5736.5	18695.5	ESTs
189	162533	0.262	5633	19119.3	Human breast tumor autoantigen mRNA, complete sequence
190	126230	0.261	2759.8	9403.3	ESTs
191	208434	0.261	5915.4	20171	ESTs
192	309893	0.259	4439.5	15220.2	Hormone receptor (growth factor-inducible nuclear protein N10)
193	39884	0.258	2625.3	9025.7	Unknown EST
194	210548	0.257	1521.3	5261.7	ESTs
195	196109	0.257	1818	6289.1	ESTs
196	293078	0.256	814.4	2827.2	ESTs
197	129616	0.248	5062.1	18165.2	ESTs
198	128245	0.242	5468.9	20059.2	ESTs
199		0.239	1411.9	5244.8	
200	685801	0.237	11696.4	43864.3	Human bumetanide-sensitive Na-K-Cl cotransporter (NKCC1) mRNA, complete cds
201	296562	0.236	3666.6	13822.4	ESTs
202		0.229	1253.2	4867.3	
203	207448	0.228	4361.6	16967.1	ESTs
204		0.227	827.9	3239.1	
205	134537	0.226	3677.9	14446.5	ESTs
206	empty	0.225	1224.8	4839.3	empty
207	296552	0.222	2173	8697.9	ESTs
208	295590	0.214	6921.8	28670	ESTs
209	281125	0.214	1987.3	8255.2	ESTs
210	214331	0.213	6458.1	26924.2	ESTs
211	127646	0.211	2769.5	11654.1	ESTs
212	208940	0.21	2744.3	11626.8	ESTs
213	197856	0.209	2680.6	11401.8	ESTs
214		0.202	3014.1	13268.2	
215	197637	0.201	3230.3	14255.6	ESTs
216	42373	0.2	1375.3	6120.6	Crystallin Mu

No.	Clone ID	Cal. Ratio	Recur. Intensity	Primary Intensity	Description
217	293421	0.197	6574.1	29716.3	ESTs
218		0.196	1094.5	4973	
219		0.195	1242.5	5655.7	
220	214043	0.191	2538.1	11804.5	ESTs
221		0.188	1109.4	5231.6	
222		0.188	2335.2	11018	
223		0.182	980.4	4781	
224		0.182	2486	12136.9	
225	196125	0.181	3092.1	15215.2	ESTs
226	897531	0.18	689.9	3404.7	Fibronectin 1
227	239711	0.18	8520.1	42094.2	ESTs
228	293457	0.179	6499.4	32266.6	ESTs
229		0.175	1180.1	5974.5	
230	293785	0.175	5652.5	28719.3	ESTs
231	210610	0.173	7021.7	35999.1	ESTs
232		0.17	3461.3	18134.2	
233	130053	0.169	2243.8	11786.8	ESTs
234		0.168	1317.8	6987.6	
235	121981	0.167	2203.3	11749.7	ESTs
236	203400	0.165	2815	15167.5	ESTs
237	230613	0.164	2593.4	14035.1	ESTs
238		0.164	1016.5	5502.8	
239		0.161	1346.2	7423.3	
240	empty	0.159	755.2	4226	empty
241	243403	0.157	2169.8	12277.6	ESTs
242	137396	0.157	3718.8	21081.7	ESTs
243	empty	0.151	2764.8	16304.7	empty
244	295044	0.148	1753.4	10522.5	ESTs
245		0.146	2720	16582	
246	211951	0.142	4429.6	27655.7	ESTs
247		0.139	3244.3	20772.1	
248	138601	0.138	2613.6	16778.8	ESTs
249	293306	0.122	2773.6	20221.8	ESTs
250	240138	0.116	3188.1	24440.5	ESTs
251	212098	0.107	3165.6	26193.8	ESTs

CLAIMS

1. A method of diagnosing or prognosing development or progression of prostate cancer in a subject, comprising detecting an abnormality in at least one HRPC-related molecule of the subject, wherein at least one such molecule is represented by Image ID Clone: 781047, 785778, 842968, 769921, 898286, 204214, 814701, 435076, 531319, 415089, 898062, 453107, 785707, 795936, 700792, 34778, 46182, 769921, 783697, 451907, 711768, 416833, 810711, 789204, 789182, 725454, 951142, 49352, 273546, 46717, 855487, 41117, 26578, 684655, 45233, 814117, 810552, 739511, 283315, 897774 or 2911545 (VDUP1).

2. The method of claim 1, comprising detecting abnormalities in more than one HRPC-related molecule.

3. The method of claim 2, wherein at least a second HRPC-related molecule is represented by a molecule listed in Table 1 or Table 4.

4. The method of claim 2, further comprising detecting an abnormality in at least one HRPC-related molecule not listed in Table 1 or Table 4.

5. The method of claim 1, comprising detecting an increase or decrease in expression or activity level of S100P, FKBP5, LMO4, CRYM, or a combination of two or more thereof.

6. A method of diagnosing or prognosing development or progression of prostate cancer in a subject, comprising detecting an abnormality in at least 5, at least 10, at least 15, at least 25, at least 50, or at least 100 HRPC-related nucleic acid molecules listed in Table 1 or Table 4 or encoded for by a nucleic acid molecule listed in Table 1 or Table 4.

7. The method of claim 2, wherein an abnormality is detected in at least 5, 10, 15, 25, 50, or 100 HRPC-related nucleic acid molecules listed in Table 1 or Table 4 or encoded for by a nucleic acid molecule listed in Table 1 or Table 4.

8. The method of claim 1 or claim 6, where an abnormality comprises over- or under-expression of the HRPC-related molecule.

9. The method of claims 8, wherein an abnormality is over-expression.

10. The method of claim 9, where at least one HRPC-related molecule is represented by Image Clone ID number: 1475595, 1460110, 50794, 78294, 190491, 66731, 143287, 754600, 754509, 308041, 70827, 361974, 503097, 796646, 41650, 841641, 724615, 839101, 504226, 810711, 435330, 773567, 431296, 345232, 756405, 256907, 415817, 366541, 223350, 366067, 724831, 814353, 236034, 809910, 1470048, 1323448, 1456424, 453689, 135221, 340734, 180864, 768562, 179276, 44505, 293104, 243343, 66317, 812251, 245920, 265874, 770212, 784910, 839094, 712049, 669435, 841470, 782339, 297061, 429466, 300137, 487172, 343744, 795730, 268876, 742132, 755578, 502682, 510381, 140574, 135630, 278242, 742862, 1049033, 270136, 768260, 53039, 211813, 195051, 125769, 122955, 129342, 292392, 139331, 143995, 139250, 243360, 194307, 235040, 295483, or 143756.

11. The method of claim 8, wherein an abnormality is underexpression.

12. The method of claim 11, where at least one HRPC-related molecule is represented by Image Clone ID number: 897768, 1456160, 34778, 810512, 753184, 200814, 470393, 23185, 128126, 42373, 511521, 810117, 950682, 783696, 815555, 897531, 713145, 502690, 469969, 309893, 725877, 343987, 49318, 42864, 193087, 162533, 1309620, 685801, 825740, 756708, 28469, 187147, 246304, 130280, 753587, 123980, 241985, 564621, 841507, 810703, 784772, 143306, 246722, 298417, 51582, 757222, 884783, 417424, 324891, 504791, 725877, 743230, 377048, 42627, 144797, 244955, 204735, 144747, 292749, 196109, 120375, 121981, 121715, 243403, 127409, 130053, 243291, 203514, 133130, 134495, 296552, 138601, 167076, 197323, 197637, 194906, 194985, 196125, 196303, 243784, 280122, 245235, 197856, 200604, 203400, 207448, 234469, 210548, 208940, 208434, 211951, 212098, 233399, 240138, 137396, 241097, 239835, 308231, 292312, 292391, 293421, 293306, 293785, 295044, 295590, 296102, 296602, 297110, 191572, 195132, 233274, 246546, 296562, 214331, 214043, 126230, 128245, 129616, 134312, 230613, 239711, 134537, 127646, 136984, 210610, 293457, 233299, 281125, 26184, 39093, or 39884.

13. The method of claim 1 or claim 6, comprising:

measuring an amount of the HRPC-related molecule in a sample derived from the subject, in which a difference in level of the HRPC-related molecule relative to that present in a sample derived from the subject at an earlier time, is diagnostic or prognostic for development or progression of prostate cancer.

14. The method of claim 1 or claim 6, wherein detecting an abnormality comprises:

measuring a HRPC-related molecule level in a sample derived from the subject, in which a difference in the HRPC-related molecule level in the sample, relative to the HRPC-related molecule level found in an analogous sample from a subject not having the disease or disorder, or a standard HRPC-related molecule level in analogous samples from a subject not having the disease or disorder or not having a predisposition developing the disease or disorder, is an abnormality in that HRPC-related molecule.

15. The method of claim 1 or claim 6, wherein detecting an abnormality comprises:

measuring a level of HRPC-related protein functional activity in a sample derived from the subject, in which a difference in the level of HRPC-related protein functional activity in the sample, relative to the level of HRPC-related protein functional activity found an analogous sample from a subject not having the disease or disorder or a standard HRPC-related protein functional activity level in analogous samples from a subject not having the disease or disorder or not having a predisposition for developing the disease or disorder, is an abnormality in that HRPC-related molecule.

16. The method of claim 1 or claim 6, where the HRPC-related molecule is a HRPC-related nucleic acid molecule (DNA or RNA or cDNA) or a HRPC-related protein.

17. The method of claim 16, wherein at least one HRPC-related molecule is a HRPC-related nucleic acid.

18. The method of claim 17, comprising *in vitro* nucleic acid amplification.

19. The method of claim 17, comprising nucleic acid hybridization.

20. The method of claim 19, further comprising determining the amount of hybridization.

21. The method of claim 16, wherein at least one HRPC-related molecule is a HRPC-related protein.

22. The method of claim 21, wherein detecting the abnormality comprises:

contacting a sample from the subject with a HRPC protein-specific binding agent; and

detecting whether the binding agent is bound by the sample and thereby measuring the levels of the HRPC-related protein present in the sample, in which a difference in the level of HRPC-related protein in the sample, relative to the level of HRPC-related protein found an analogous sample from a subject not having the disease or disorder, or a standard HRPC-related protein level in analogous samples from a subject not having the disease or disorder or not having a predisposition for developing the disease or disorder, is an abnormality in that HRPC-related molecule.

23. The method according to claim 23, wherein the specific binding agent is detectably labeled.

24. The method of claim 1 or claim 6, wherein the abnormality is detected in a sample from the subject, and the sample comprises serum.

25. The method of claim 1 or claim 6, wherein the abnormality is detected in a sample from the subject, and the sample comprises prostate tissue.

26. The method of claim 17, comprising:

providing nucleic acids from the subject;

amplifying the nucleic acids to form nucleic acid amplification products;

contacting the nucleic acid amplification products with an oligonucleotide probe that will hybridize under stringent conditions with a nucleic acid encoding a HRPC-related protein;

detecting the nucleic acid amplification products which hybridize with the probe; and

quantifying the amount of the nucleic acid amplification products that hybridize with the probe.

27. The method of claim 26, where the sequence of the oligonucleotide probe is selected to bind specifically to a nucleic acid molecule listed in Table 1 or Table 4.

28. The method of claim 26, where the primers are selected to amplify a nucleic acid molecule listed in Table 1.

29. The method of claim 26, where the primers are selected to amplify a nucleic acid product encoding cartilage glycoprotein-39 (CHI3L1), S-100P PROTEIN (S100P), CX3C chemokine/fractalkine (SCYD1), adenylate kinase 1 (AK1), forkhead transcription factor HFH-4 (HFH-4) (FKHL13), UDP glucuronosyltransferase precursor (UGT2B15), Pleiotrophin (heparin binding growth factor 8) (PTN), heat shock 27kD protein 2/Alpha-B-crystallin (HSP27), Proteasome (prosome, macropain) subunit, beta type, 5 (PSMB5), Inhibitor of NFKB (NFKBIA), interferon-induced 17 kD protein (ISG15), MAP kinase activated protein kinase 2 (MAPKAPK2), signal transduction protein (SH3 containing) (EFS2), hkf-1 Zinc finger protein (ZFP103), chromosome

condensation 1 (CHC1), CDP-diacylglycerol synthase (CDS1), gap junction protein, alpha 1, 43kD (connexin 43) (GJA1), cyclin D1 (CCND1), Inhibitor of DNA binding 3, helix-loop-helix protein (ID3), H1 histone family, member2 (H1F2), Cytochrome B561 (CYB561), Cathepsin H (CTSH), calcineurin alpha (PPP3CA), 54 kDa progesterone receptor-associated immunophilin (FKBP5), translocation protein 1 (TLOC1), Clusterin (complement lysis inhibitor; testosterone-repressed prostate message 2; apolipoprotein J) (CLU), Pulmonary surfactant-associated protein A (SFTPA1), protease inhibitor 12 (PI12; neuroserpin) (PI12), Thrombospondin 1 (THBS1), Ribophorin I (RPN1), A disintegrin-like and metalloprotease (reprolysin type) with thrombospondin type 1 motif, 1 (ADAMTS1), Collagen, type IV, alpha 5 (Alport syndrome) (COL4A5), LIM domain only 4 / breast tumor autoantigen (LMO4), bumetanide-sensitive Na-K-Cl cotransporter (NKCC1) (SLC12A2), Fibronectin (FN1), Crystallin Mu (CRYM) or "upregulated by 1,25-dihydroxyvitamin D-3" (VDUP1).

30. The method of claim 1 or claim 6, comprising detecting a mutation, duplication or deletion of a HRPC-related nucleic acid in cells of the individual.

31. The method of claim 1 or claim 6, comprising detecting decreased, increased, or mutant HRPC-related protein in cells of the individual.

32. A method of selecting a prostate cancer therapy, comprising:

detecting an abnormality in at least one HRPC-related molecule of a subject; and

if such abnormality is identified, selecting a treatment to prevent or reduce hormone-refractory prostate cancer or to delay the onset of hormone-refractory prostate cancer.

33. The method of claim 32, wherein the at least one HRPC-related molecule is SCYD1, S100P, CCND1, CRIP1, ISG15, SCNN1A, ZFP103, MAPKAPK2, UTG2B15, RABGGTA, NFKBIA, SLCYA5, AP3B2, PTPN2, FOXJ1, APOC1, FLJ23538, OXCT, PFKP, TNRC3, HXB, PFKP, OAT, PFKP, RFP, THBS1, LMO4, MLD, CRYM, MME, HMGCS2, SLC12A2, ODC1, EIF4EBP1, CDS1, FKBP4, VDUP1, or FKBP5.

34. The method of claim 32, comprising detecting an abnormality in more than one HRPC-related molecule of a subject, wherein the more than one HRPC-related molecules are two or more of SCYD1, S100P, CCND1, CRIP1, ISG15, SCNN1A, ZFP103, MAPKAPK2, UTG2B15, RABGGTA, NFKBIA, SLCYA5, AP3B2, PTPN2, FOXJ1, APOC1, FLJ23538, OXCT, PFKP, TNRC3, HXB, PFKP, OAT, PFKP, RFP, THBS1, LMO4, MLD, CRYM, MME, HMGCS2, SLC12A2, ODC1, EIF4EBP1, CDS1, FKBP4, VDUP1, or FKBP5.

35. The method of claim 32, wherein the at least one HRPC-related molecule is S100P, FKBP5, LMO4, CRYM, or a combination of two or more thereof.

36. The method of claim 32, further comprising treating the subject with the selected treatment.

37. The method of claim 36, wherein the selected treatment comprises treating the subject with rapamycin, or a derivative, mimetic, or analog of rapamycin.

38. The method of claim 32, wherein detecting an abnormality in at least one HRPC-related molecule of a subject comprises quantitatively or qualitatively analyzing a DNA, mRNA, cDNA, protein, or protein modification.

39. A method of modifying a level of expression of a HRPC-related protein in a subject, comprising:

expressing in the subject a recombinant genetic construct comprising a promoter operably linked to a nucleic acid molecule, wherein the nucleic acid molecule comprises at least 10 consecutive nucleotides of a HRPC-related nucleic acid sequence, wherein expression of the nucleic acid molecule changes expression of the HRPC-related protein, and wherein the HRPC-related nucleic acid sequence is represented by Image ID Clone number 781047, 785778, 842968, 769921, 898286, 204214, 814701, 435076, 531319, 415089, 898062, 453107, 785707, 795936, 700792, 34778, 46182, 769921, 783697, 451907, 711768, 416833, 810711, 789204, 789182, 725454, 951142, 49352, 273546, 46717, 855487, 41117, 26578, 684655, 45233, 814117, 810552, 739511, 283315, or 897774.

40. The method of claim 39 wherein the nucleic acid molecule is in antisense orientation relative to the promoter.

41. The method of claim 39 wherein the nucleic acid molecule is in sense orientation relative to the promoter.

42. The method of claim 39, wherein the recombinant genetic construct expresses a siRNA corresponding to a HRPC-related nucleic acid sequence.

43. A kit for measuring a HRPC-related molecule level, comprising a binding molecule that selectively binds to the HRPC-related molecule, wherein the HRPC-related molecule is represented by Image ID Clone number 781047, 785778, 842968, 769921, 898286, 204214, 814701, 435076, 531319, 415089, 898062, 453107, 785707, 795936, 700792, 34778, 46182, 769921, 783697, 451907, 711768, 416833, 810711, 789204, 789182, 725454, 951142, 49352, 273546, 46717, 855487, 41117, 26578, 684655, 45233, 814117, 810552, 739511, 283315, or 897774.

44. The kit of claim 43, wherein the levels of a plurality of HRPC-related molecules are measured.

45. The kit of claim 44, comprising an array.

46. The kit of claim 43, wherein the HRPC-related molecule level is a HRPC-related protein level, and the binding molecule is an antibody or antibody fragment that selectively binds a HRPC-related protein.

47. The kit of claim 43, wherein the HRPC-related molecule level is a HRPC-related nucleic acid molecule level, and the binding molecule is an oligonucleotide capable of hybridizing to the HRPC-related nucleic acid molecule.

48. The method of claim 1 or claim 6, wherein detecting the abnormality comprises:

determining whether a HRPC-related gene expression profile from the subject indicates development or progression of prostate cancer.

49. The method of claim 48, wherein the gene expression profile comprises an array.

50. The method of claim 48, comprising:

comparing the HRPC-related gene expression profile from the subject to at least one control gene expression fingerprint for a specific stage of prostate cancer.

51. The method of claim 50, where the at least one control gene expression profile is a fingerprint for a normal prostate tissue, a primary prostate cancer tissue, a prostate cancer tissue responding to androgen ablation therapy, or a hormone refractory prostate cancer tissue.

52. A method of screening for a compound useful in treating, reducing, or preventing prostate cancer or development or progression of prostate cancer, comprising determining if application of a test compound alters a HRPC-related gene expression profile so that the profile more closely resembles a prostate-linked profile than it did prior to such treatment, and selecting a compound that so alters the HRPC-related gene expression profile, wherein the HRPC-related gene expression profile includes at least one molecule represented by Image ID Clone number 781047, 785778, 842968, 769921, 898286, 204214, 814701, 435076, 531319, 415089, 898062, 453107, 785707, 795936, 700792, 34778, 46182, 769921, 783697, 451907, 711768, 416833, 810711, 789204, 789182, 725454, 951142, 49352, 273546, 46717, 855487, 41117, 26578, 684655, 45233, 814117, 810552, 739511, 283315, 897774, or 2911545.

53. The method of claim 52, wherein the compound inhibits or inactivates a molecule represented by those listed in Table 1 or Table 4.

54. The method of claim 52, wherein the test compound is applied to a test cell.

55. The method of claim 52, comprising:

contacting test cells with a test compound; and

measuring at least one HRPC-related molecule level and/or activity in the test cells, in which a difference in HRPC-related molecule level and/or activity in the test cells, relative to the analogous HRPC-related molecule level and/or activity found in analogous cells not contacted with the test compound, indicates that the test compound is useful in treating, reducing, or preventing prostate cancer or development or progression of prostate cancer.

56. The method of claim 55, wherein at least one HRPC-related molecule is a nucleic acid molecule listed in Table 1 or Table 4, or is encoded for by a nucleic acid molecule listed in Table 1 or Table 4.

57. The method of claim 55, in which measuring at least one HRPC-related molecule level and/or activity comprises:

creating a HRPC-related gene expression profile for the test cell after contacting the cell with the test compound; and

comparing the test cell HRPC-related gene expression profile to at least one control gene expression profile for a specific stage of prostate cancer.

58. The method of claim 57, where the control gene expression profile is a profile for a normal prostate tissue, a primary prostate cancer tissue, a prostate cancer tissue responding to androgen ablation therapy, or a hormone refractory prostate cancer tissue.

59. The method of claim 52, wherein the profile comprises an array.

60. A compound selected by the method of claim 52.

61. The method of claim 36, wherein the selected treatment comprises treating the subject with FR901464, or a derivative, mimetic, or analog of FR901464.

FIG 1A

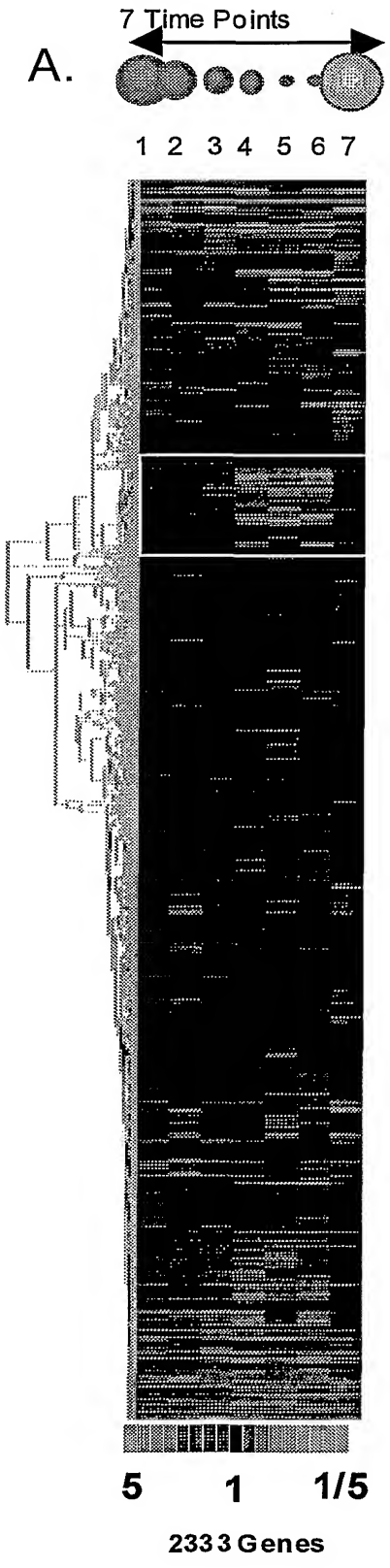


FIG 1B

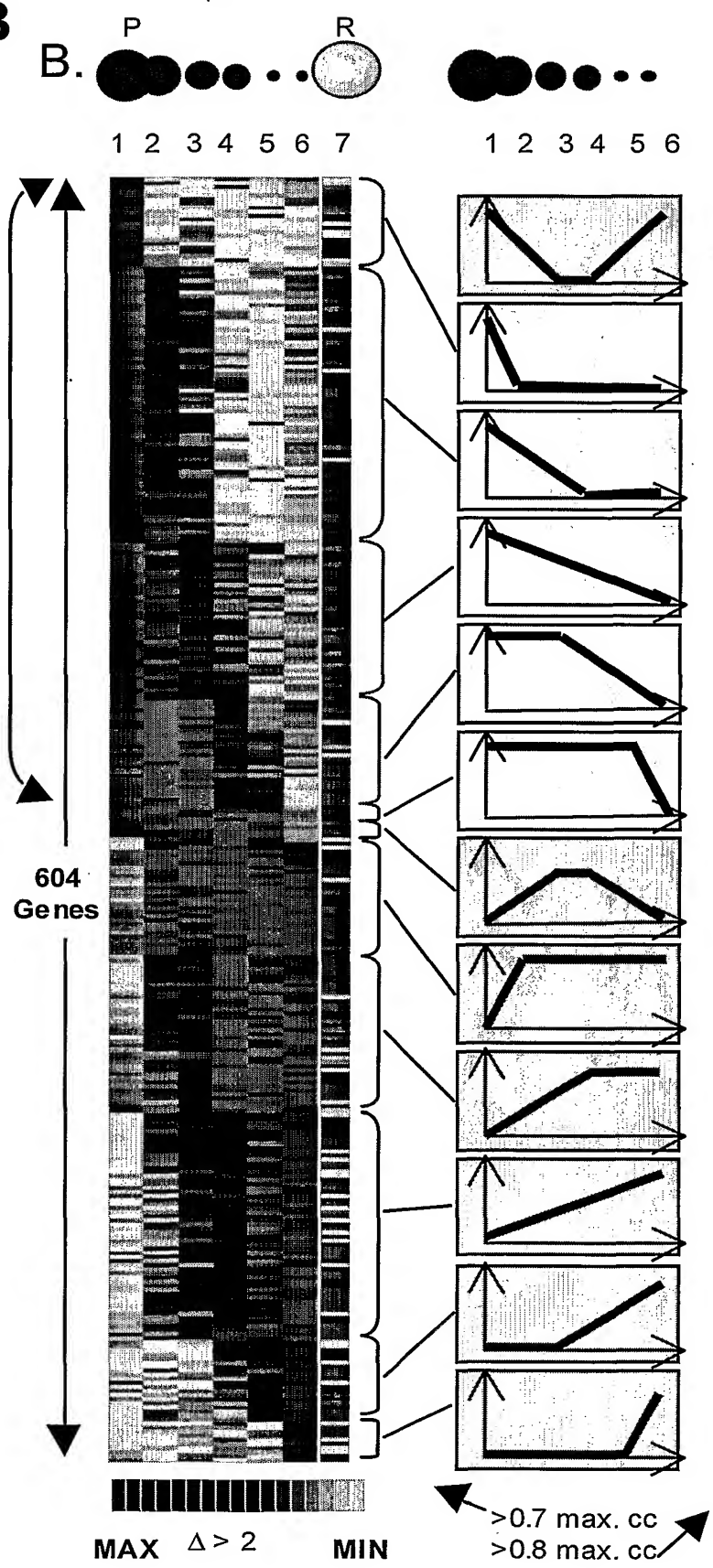
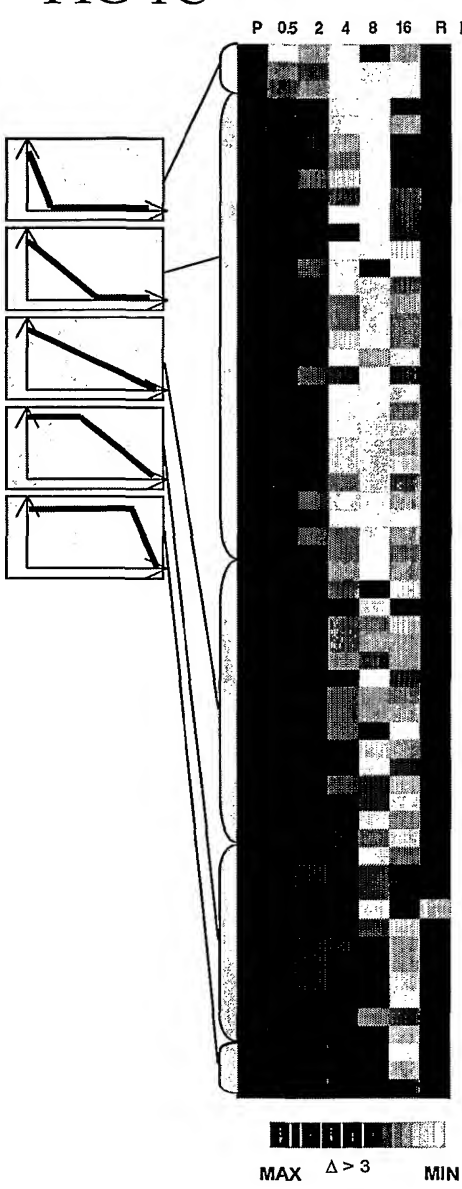
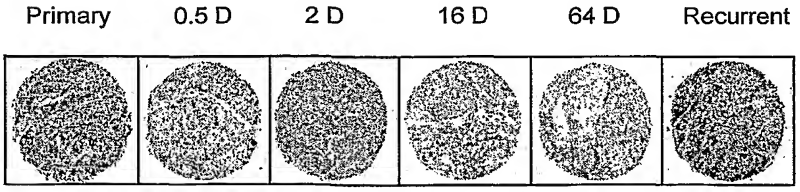


FIG 1C



Fold Δ		IMAGE Clone ID	
3.8	ISG15	742132	interferon-stimulated protein, 15 kDa
3.1		767817	H.Sapiens gene for RNA polymerase II subunit
7.1	TM4SF1	840567	transmembrane 4 superfamily member 1
3.0	BUB3	785778	Homo sapiens mRNA for 6.2 kd protein
3.7	PGAM2	283315	phosphoglycerate mutase 2 (muscle)
4.0		814117	ESTs
3.0	CCT6	45233	chaperonin containing T-complex subunit 6
3.1		29063	Homo sapiens clone 23620 mRNA sequence
3.2	PSMC2	684655	proteasome (prosome, macropain) 26S subunit,
3.7		134719	ESTs
3.2	CAP	24145	adenylyl cyclase-associated protein
3.4	CDC20	898062	cell division cycle 20, S.cerevisiae homolog
5.1	TCRG	281003	T-cell receptor, gamma cluster
3.5	ASAH	855487	N-acylsphingosine amidohydrolase (acid
3.4	DLD	42059	dihydrolipoamide dehydrogenase (E3
3.3	MDH1	53316	malate dehydrogenase 1, NAD (soluble)
3.3		273546	H.sapiens ADE2H1 mRNA showing
4.1		293727	ESTs
3.0	ANX7	49352	annexin VII (synexin)
6.2	CKS2	725454	CDC28 protein kinase 2
5.1		856427	Homo sapiens HPV16 E1 protein binding protein
3.1	PCNA	789182	proliferating cell nuclear antigen
7.0	STK12	531319	serine/threonine kinase 12
4.0	AMD1	149013	S-adenosylmethionine decarboxylase 1
3.4	TUBA3	47833	Tubulin, alpha, brain-specific
3.3		626716	ESTs
3.5	TLOC1	789204	translocation protein 1
6.3	FECH	66729	*ferrochelatase (protoporphyria)*
3.9	MLLT7	509887	myeloid/lymphoid or mixed-lineage leukemia
3.1	CDC45L	453107	CDC45 (cell division cycle 45, S.cerevisiae,
3.7	FKBP5	416833	FK506-binding protein 5
3.1		711768	ESTs
3.1		451907	Homo sapiens ZW10 interactor Zwint mRNA,
5.1		66406	ESTs
4.7		129865	ESTs
3.8	ODC1	796846	ornithine decarboxylase 1
4.7	CDC18L	204214	CDC18 (cell division cycle 18, S.pombe,
8.5	CDC2	898266	Homo sapiens mRNA for CDC2 delta T.
5.5	MAD2L1	814701	MAD2 (mitotic arrest deficient, yeast,
5.8	FKBP5	416833	FK506-binding protein 5
3.5	LDHA	43550	lactate dehydrogenase A
5.2	CENPF	435076	centromere protein F (400kD)
3.5		207358	Human (HepG2) glucose transporter gene
3.1		249603	ESTs
5.2	UBCH10	769921	Human cyclin-selective ubiquitin carrier protein
3.2	ADAM9	204257	a disintegrin and metalloproteinase domain
8.1	INSIG1	207288	insulin induced gene 1
4.8	FOS	811015	v-fos FBJ murine osteosarcoma viral
3.7	MLD	324891	membrane fatty acid (lipid) desaturase
5.1	CDKN3	700792	cyclin-dependent kinase inhibitor 3
4.0		131316	ESTs
3.6	TSN	795936	translin
3.7		126650	ESTs
4.1	BUB1	781047	budding uninhibited by benzimidazoles 1 (yeast
3.4	ADM	774446	adrenomedullin
4.5		785707	ESTs, Highly similar to protein regulating
4.5	NEK2	415089	NIMA (never in mitosis gene a)-related kinas
4.0		196303	ESTs
4.4		788256	ESTs

FIG 1D



Ki67 IHC

FIG 2

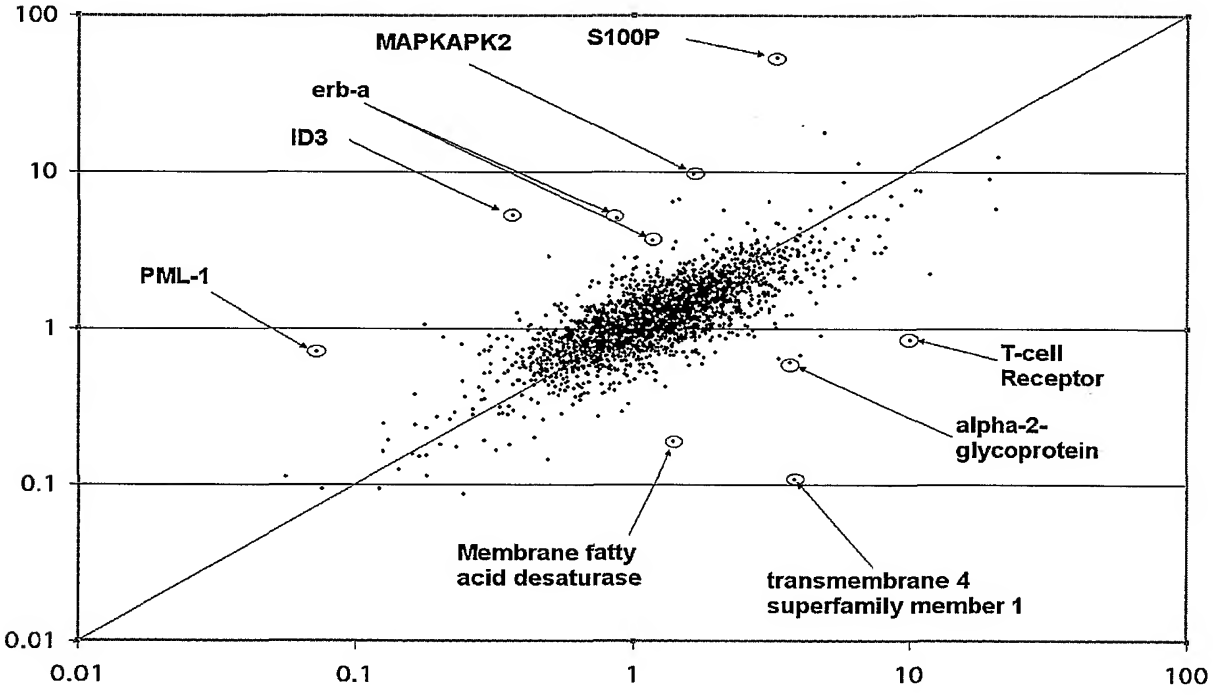
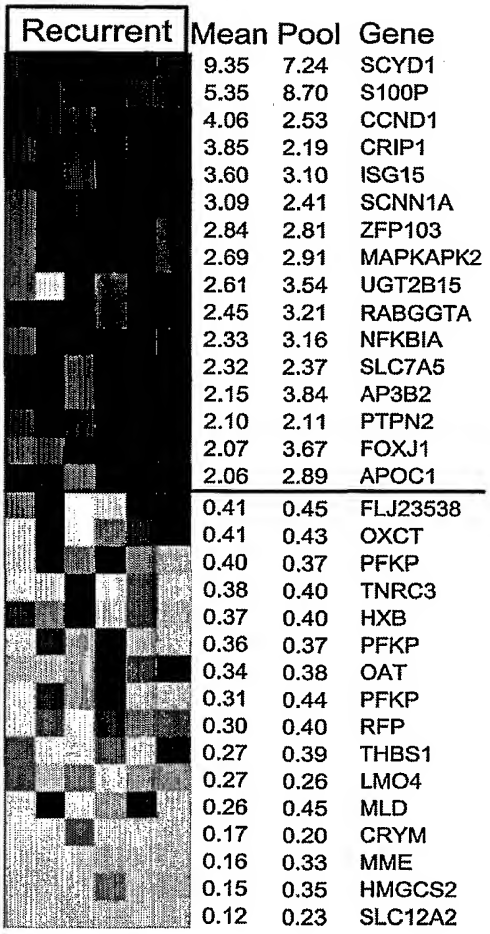

100
10
1
0.1
0.01
0.01
0.1
1
10
100
MAPKAPK2
S100P
erb-a
ID3
PML-1
T-cell
Receptor
alpha-2-
glycoprotein
Membrane fatty
acid desaturase
transmembrane 4
superfamily member 1

FIG 3A



Recurrent	Mean	Pool	Gene
Max. 28.01	9.35	7.24	SCYD1
17.72	5.35	8.70	S100P
13.04	4.06	2.53	CCND1
8.63	3.85	2.19	CRIP1
9.68	3.60	3.10	ISG15
5.76	3.09	2.41	SCNN1A
6.96	2.84	2.81	ZFP103
6.33	2.69	2.91	MAPKAPK2
7.89	2.61	3.54	UGT2B15
8.63	2.45	3.21	RABGGTA
4.59	2.33	3.16	NFKBIA
6.66	2.32	2.37	SLC7A5
8.19	2.15	3.84	AP3B2
3.37	2.10	2.11	PTPN2
5.46	2.07	3.67	FOXJ1
5.12	2.06	2.89	APOC1
Min. 0.17	0.41	0.45	FLJ23538
0.16	0.41	0.43	OXCT
0.22	0.40	0.37	PFKP
0.09	0.38	0.40	TNRC3
0.22	0.37	0.40	HXB
0.13	0.36	0.37	PFKP
0.23	0.34	0.38	OAT
0.09	0.31	0.44	PFKP
0.16	0.30	0.40	RFP
0.13	0.27	0.39	THBS1
0.18	0.27	0.26	LMO4
0.05	0.26	0.45	MLD
0.09	0.17	0.20	CRYM
0.09	0.16	0.33	MME
0.09	0.15	0.35	HMGCS2
0.04	0.12	0.23	SLC12A2

FIG 3B

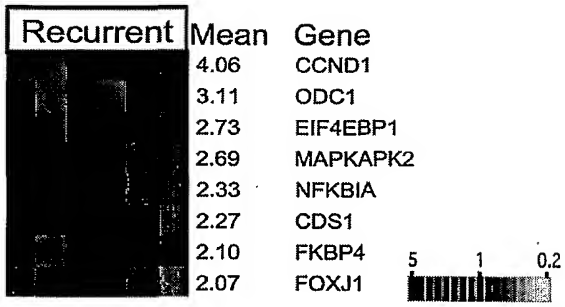
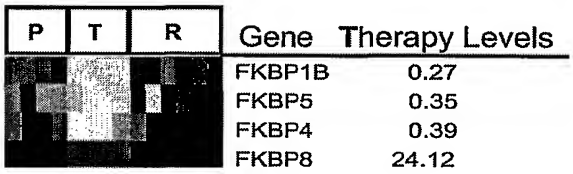


FIG 3C



P	T	R	Gene	Therapy Levels
			FKBP1B	0.27
			FKBP5	0.35
			FKBP4	0.39
			FKBP8	24.12

FIG 3D

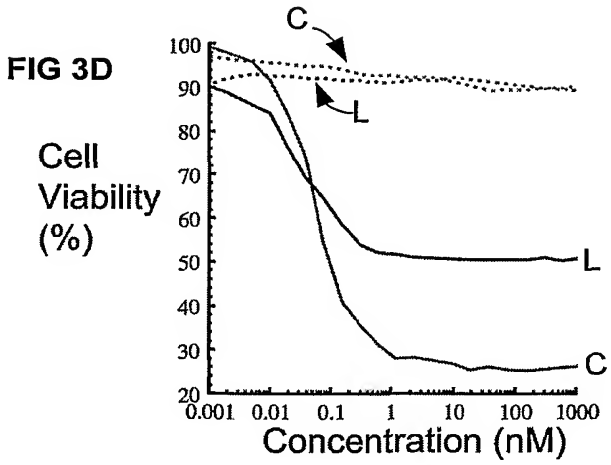


FIG 4

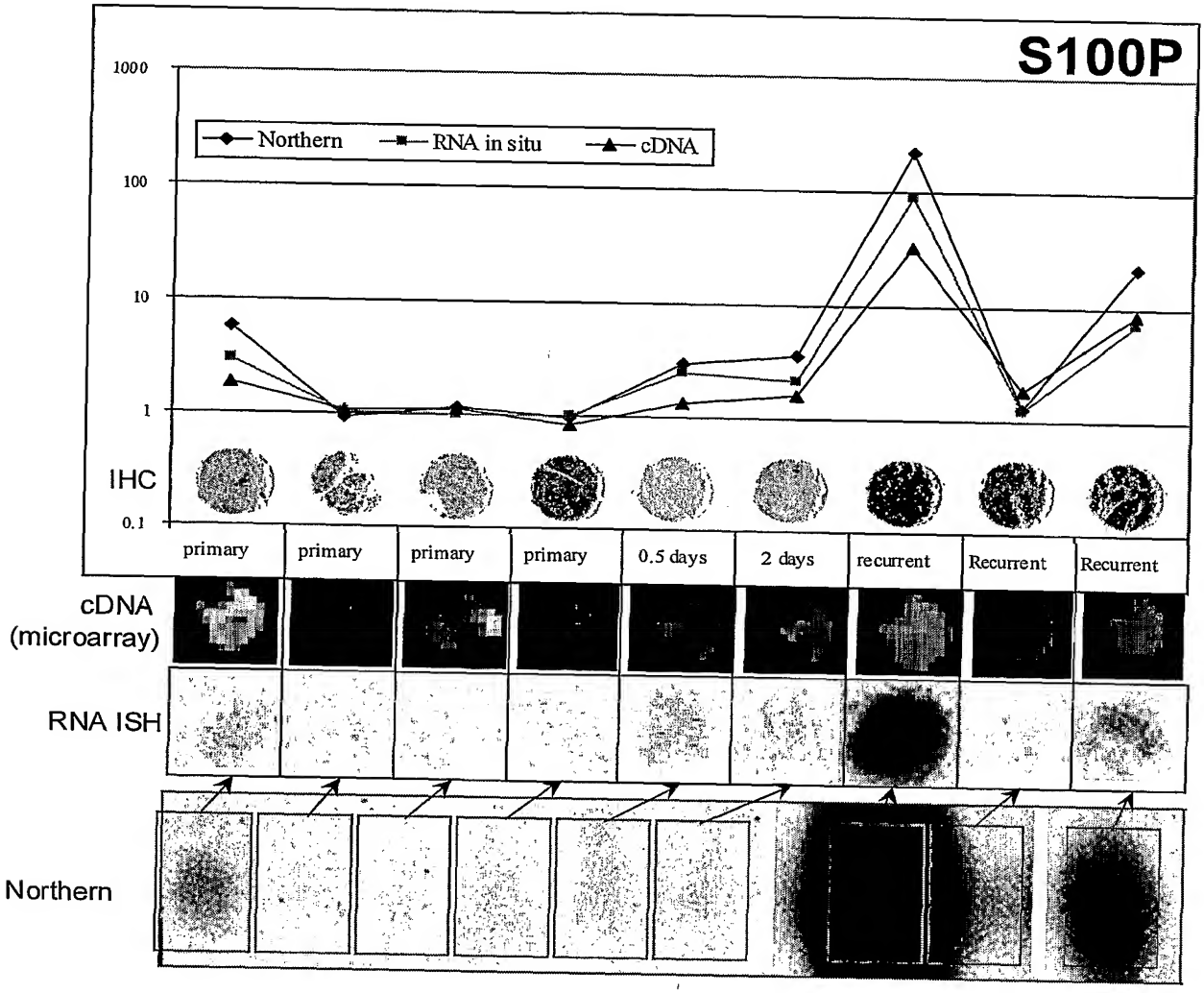

S100P
1000
100
10
1
0.1
Northern
RNA in situ
cDNA
IHC
primary
primary
primary
primary
0.5 days
2 days
recurrent
Recurrent
Recurrent
cDNA (microarray)
RNA ISH
Northern

FIG 5

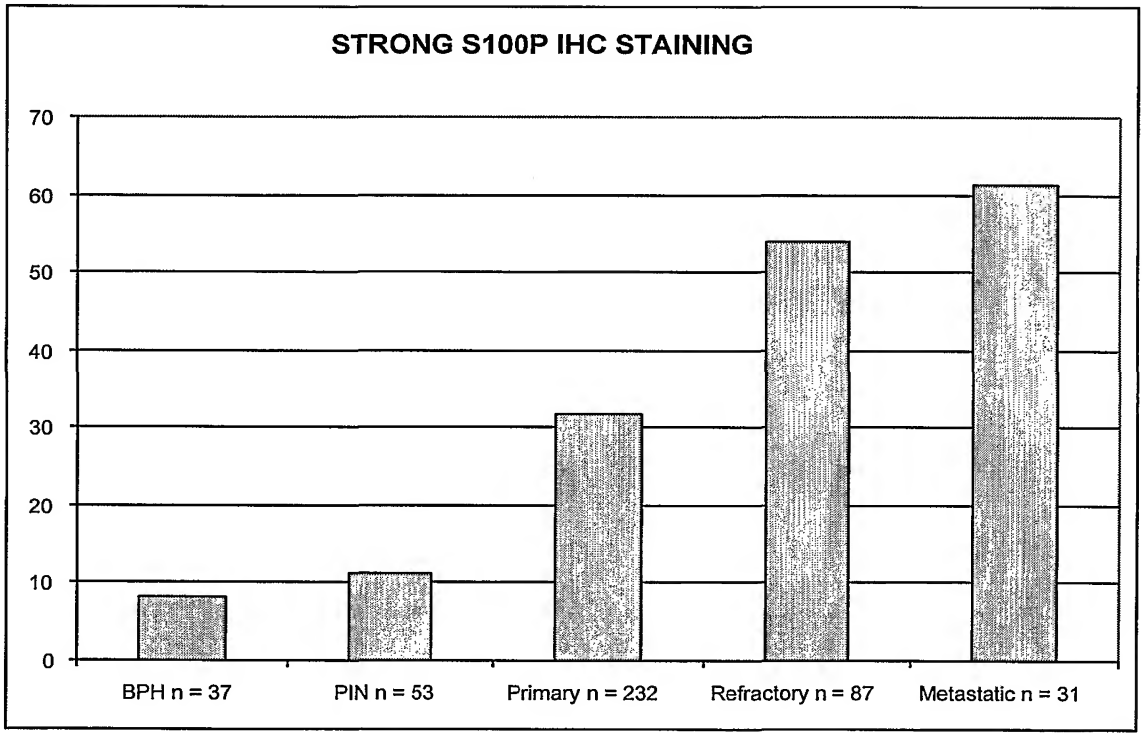

STRONG S100P IHC STAINING
70
60
50
40
30
20
10
0
BPH n = 37
PIN n = 53
Primary n = 232
Refractory n = 87
Metastatic n = 31

FIG 6A

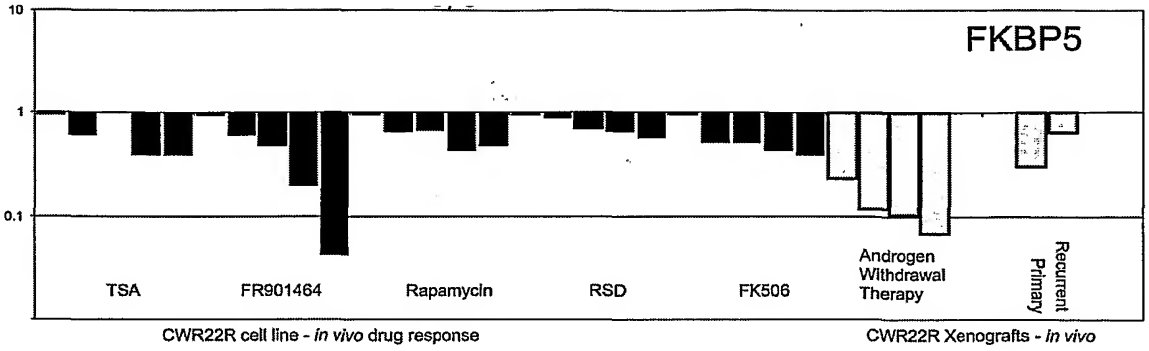


FIG 6B

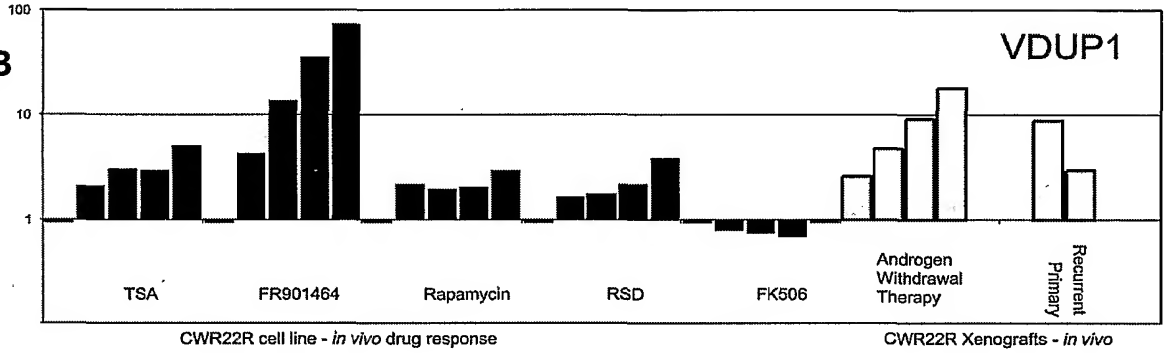


FIG 6C

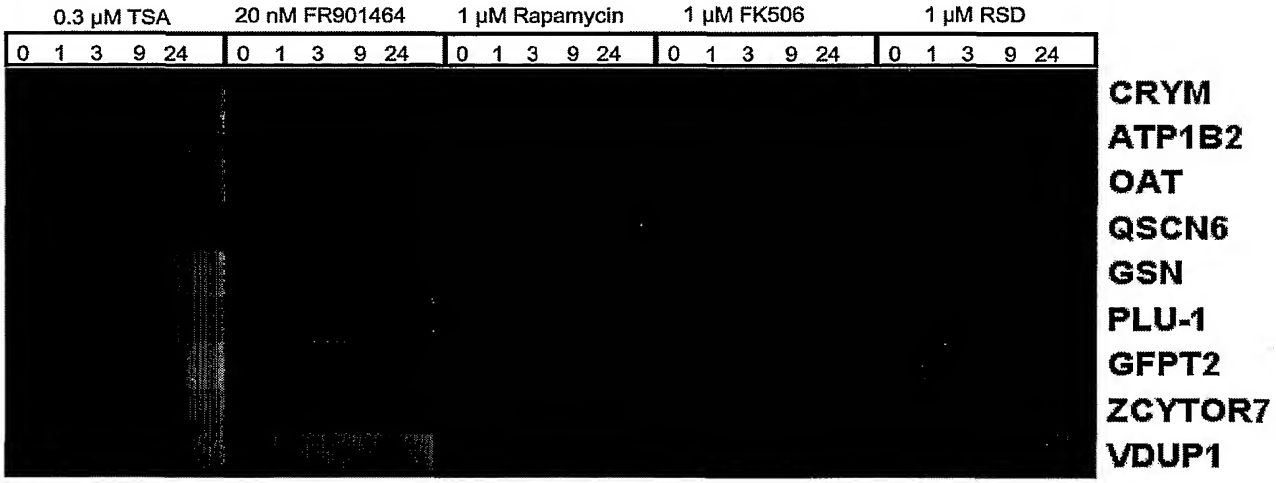
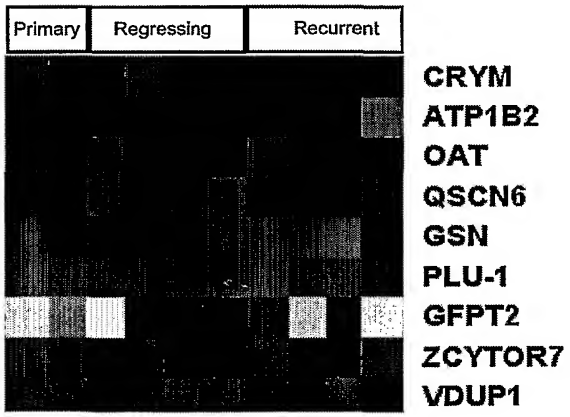


FIG 6D



SEQUENCE LISTING

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STATEMENT ACCOMPANYING SEQUENCE LISTING

The sequence listing does not include matter that goes beyond the disclosure in the international application.

The printout of the attached Sequence Listing is identical to the computer readable sequence listing on the enclosed computer disk.